```
=> file biosis caba caplus embase japio lifesci medline scisearch
=> e liu jun/au
E1            2     LIU JUMMING/AU
E2            1     LIU JUMN HUA/AU
E3         5804 --> LIU JUN/AU
E4           17     LIU JUN AN/AU
E5            2     LIU JUN ANG/AU
E6            6     LIU JUN BAO/AU
E7            4     LIU JUN BIN/AU
E8            2     LIU JUN BING/AU
E9           28     LIU JUN BO/AU
E10           1     LIU JUN CHANG/AU
E11           6     LIU JUN CHAO/AU
E12           1     LIU JUN CHEN/AU

=> s e3-e12 and tuberculosis
L1           74 ("LIU JUN"/AU OR "LIU JUN AN"/AU OR "LIU JUN ANG"/AU OR "LIU
               JUN BAO"/AU OR "LIU JUN BIN"/AU OR "LIU JUN BING"/AU OR "LIU
               JUN BO"/AU OR "LIU JUN CHANG"/AU OR "LIU JUN CHAO"/AU OR "LIU
               JUN CHEN"/AU) AND TUBERCULOSIS

=> dup rem l1
PROCESSING COMPLETED FOR L1
L2            26 DUP REM L1 (48 DUPLICATES REMOVED)

=> s l2 and ((alanine dehydrogenase)or(glutamine synthetase)or(serine
dehydratase))
L3            2 L2 AND ((ALANINE DEHYDROGENASE) OR(GLUTAMINE SYNTHETASE) OR(SER
               INE DEHYDRATASE))

=> d 1-
YOU HAVE REQUESTED DATA FROM 2 ANSWERS - CONTINUE? Y/(N):y

L3   ANSWER 1 OF 2  BIOSIS  COPYRIGHT (c) 2009 The Thomson Corporation  on STN
AN   2003:127824  BIOSIS <<LOGINID::20090416>>
DN   PREV200300127824
TI   Mycobacterium bovis BCG vaccines exhibit defects in alanine and serine
     catabolism.
AU   Chen, Jeffrey M.; Alexander, David C.; Behr, Marcel A.;   ***Liu, Jun***
     [Reprint Author]
CS   Department of Medical Genetics and Microbiology, University of Toronto, 1
     King's College Circle, 4382 Medical Sciences Building, Toronto, ON, M5S
     1A8, Canada
     jun.liu@utoronto.ca
SO   Infection and Immunity, (February 2003) Vol. 71, No. 2, pp. 708-716.
     print.
     ISSN: 0019-9567 (ISSN print).
DT   Article
LA   English
ED   Entered STN: 5 Mar 2003
     Last Updated on STN: 5 Mar 2003

L3   ANSWER 2 OF 2  CAPLUS  COPYRIGHT 2009 ACS on STN
AN   2003:855955  CAPLUS <<LOGINID::20090416>>
DN   139:363579
TI     ***Tuberculosis***   vaccines including recombinant Mycobacterium
     bovis-BCG strains expressing   ***alanine***     ***dehydrogenase***  ,
```

```
       ***serine***     ***dehydratase***   and/or   ***glutamine***
       ***synthetase***
IN     ***Liu, Jun***  ; Chen, Jeffrey; Alexander, David
PA   Can.
SO   PCT Int. Appl., 78 pp.
     CODEN: PIXXD2
DT   Patent
LA   English
FAN.CNT 1
     PATENT NO.          KIND   DATE        APPLICATION NO.        DATE
     ---------------     ----   --------    --------------------   --------
PI   WO 2003089462        A2    20031030    WO 2003-CA566          20030416
     WO 2003089462        A3    20040521
         W:  AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN,
             CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH,
             GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR,
             LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM,
             PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT,
             TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW
         RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY,
             KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES,
             FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR,
             BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG
     CA 2481108           A1    20031030    CA 2003-2481108        20030416
     AU 2003218838        A1    20031103    AU 2003-218838         20030416
     GB 2403477           A     20050105    GB 2004-25165          20030416
     GB 2403477           B     20060823
     CN 1703513           A     20051130    CN 2003-802276         20030416
     JP 2006508633        T     20060316    JP 2003-586182         20030416
     JP 4233458           B2    20090304
     RU 2339692           C2    20081127    RU 2004-133751         20030416
     ZA 2004008344        A     20050907    ZA 2004-8344           20041014
     US 20070264286       A1    20071115    US 2006-511718         20060728
PRAI US 2002-372450P      P     20020416
     WO 2003-CA566        W     20030416
RE.CNT  3     THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD
              ALL CITATIONS AVAILABLE IN THE RE FORMAT


=> e chen jeffrey/au
E1           1     CHEN JEFFERY J/AU
E2           3     CHEN JEFFERY K/AU
E3          50 --> CHEN JEFFREY/AU
E4          25     CHEN JEFFREY C/AU
E5           1     CHEN JEFFREY CHAO NAN/AU
E6           1     CHEN JEFFREY CHUANG FEI/AU
E7           5     CHEN JEFFREY E/AU
E8           8     CHEN JEFFREY E K/AU
E9           1     CHEN JEFFREY F/AU
E10          4     CHEN JEFFREY H/AU
E11         20     CHEN JEFFREY J/AU
E12          1     CHEN JEFFREY JIAN/AU

=> s e1-e12 and tuberculosis
L4           1 ("CHEN JEFFERY J"/AU OR "CHEN JEFFERY K"/AU OR "CHEN JEFFREY"/AU
               OR "CHEN JEFFREY C"/AU OR "CHEN JEFFREY CHAO NAN"/AU OR "CHEN
              JEFFREY CHUANG FEI"/AU OR "CHEN JEFFREY E"/AU OR "CHEN JEFFREY
```

```
              E K"/AU OR "CHEN JEFFREY F"/AU OR "CHEN JEFFREY H"/AU OR "CHEN
              JEFFREY J"/AU OR "CHEN JEFFREY JIAN"/AU) AND TUBERCULOSIS

=> d

L4   ANSWER 1 OF 1  CAPLUS  COPYRIGHT 2009 ACS on STN
AN   2003:855955  CAPLUS <<LOGINID::20090416>>
DN   139:363579
TI     ***Tuberculosis***   vaccines including recombinant Mycobacterium
     bovis-BCG strains expressing alanine dehydrogenase, serine dehydratase
     and/or glutamine synthetase
IN   Liu, Jun;   ***Chen, Jeffrey***  ; Alexander, David
PA   Can.
SO   PCT Int. Appl., 78 pp.
     CODEN: PIXXD2
DT   Patent
LA   English
FAN.CNT 1
     PATENT NO.          KIND   DATE        APPLICATION NO.        DATE
     ---------------     ----   --------    --------------------   --------
PI   WO 2003089462        A2    20031030    WO 2003-CA566          20030416
     WO 2003089462        A3    20040521
         W:  AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN,
             CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH,
             GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR,
             LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM,
             PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT,
             TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW
         RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY,
             KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES,
             FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR,
             BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG
     CA 2481108           A1    20031030    CA 2003-2481108        20030416
     AU 2003218838        A1    20031103    AU 2003-218838         20030416
     GB 2403477           A     20050105    GB 2004-25165          20030416
     GB 2403477           B     20060823
     CN 1703513           A     20051130    CN 2003-802276         20030416
     JP 2006508633        T     20060316    JP 2003-586182         20030416
     JP 4233458           B2    20090304
     RU 2339692           C2    20081127    RU 2004-133751         20030416
     ZA 2004008344        A     20050907    ZA 2004-8344           20041014
     US 20070264286       A1    20071115    US 2006-511718         20060728
PRAI US 2002-372450P      P     20020416
     WO 2003-CA566        W     20030416
RE.CNT  3     THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD
              ALL CITATIONS AVAILABLE IN THE RE FORMAT


=> e alexander david/au
E1            2     ALEXANDER DAVE B/AU
E2           11     ALEXANDER DAVE M/AU
E3          125 --> ALEXANDER DAVID/AU
E4           53     ALEXANDER DAVID A/AU
E5            6     ALEXANDER DAVID A PROF/AU
E6            1     ALEXANDER DAVID ALAN/AU
E7            1     ALEXANDER DAVID ALAN PROF/AU
E8            1     ALEXANDER DAVID ALLEN/AU
```

```
E9            1     ALEXANDER DAVID ANDREW/AU
E10           1     ALEXANDER DAVID AUSTIN/AU
E11          53     ALEXANDER DAVID B/AU
E12           1     ALEXANDER DAVID BEDELL/AU

=> s e1-e12 and tuberculosis
L5            8 ("ALEXANDER DAVE B"/AU OR "ALEXANDER DAVE M"/AU OR "ALEXANDER
                DAVID"/AU OR "ALEXANDER DAVID A"/AU OR "ALEXANDER DAVID A PROF"/
                AU OR "ALEXANDER DAVID ALAN"/AU OR "ALEXANDER DAVID ALAN PROF"/A
                U OR "ALEXANDER DAVID ALLEN"/AU OR "ALEXANDER DAVID ANDREW"/AU
                OR "ALEXANDER DAVID AUSTIN"/AU OR "ALEXANDER DAVID B"/AU OR "ALE
                XANDER DAVID BEDELL"/AU) AND TUBERCULOSIS

=> dup rem l5
PROCESSING COMPLETED FOR L5
L6            4 DUP REM L5 (4 DUPLICATES REMOVED)

=> s l6 and ((alanine dehydrogenase)or(glutamine synthetase)or(serine
dehydratase))
L7            1 L6 AND ((ALANINE DEHYDROGENASE) OR(GLUTAMINE SYNTHETASE) OR(SER
                INE DEHYDRATASE))

=> d

L7   ANSWER 1 OF 1  CAPLUS  COPYRIGHT 2009 ACS on STN
AN   2003:855955  CAPLUS <<LOGINID::20090416>>
DN   139:363579
TI     ***Tuberculosis***   vaccines including recombinant Mycobacterium
     bovis-BCG strains expressing   ***alanine***     ***dehydrogenase***  ,
       ***serine***     ***dehydratase***   and/or   ***glutamine***
       ***synthetase***
IN   Liu, Jun; Chen, Jeffrey;   ***Alexander, David***
PA   Can.
SO   PCT Int. Appl., 78 pp.
     CODEN: PIXXD2
DT   Patent
LA   English
FAN.CNT 1
     PATENT NO.          KIND   DATE        APPLICATION NO.        DATE
     ---------------     ----   --------    --------------------   --------
PI   WO 2003089462        A2    20031030    WO 2003-CA566          20030416
     WO 2003089462        A3    20040521
         W:  AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN,
             CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH,
             GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR,
             LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM,
             PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT,
             TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW
         RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY,
             KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES,
             FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR,
             BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG
     CA 2481108           A1    20031030    CA 2003-2481108        20030416
     AU 2003218838        A1    20031103    AU 2003-218838         20030416
     GB 2403477           A     20050105    GB 2004-25165          20030416
     GB 2403477           B     20060823
     CN 1703513           A     20051130    CN 2003-802276         20030416
```

```
     JP 2006508633          T     20060316    JP 2003-586182          20030416
     JP 4233458             B2    20090304
     RU 2339692             C2    20081127    RU 2004-133751          20030416
     ZA 2004008344          A     20050907    ZA 2004-8344            20041014
     US 20070264286         A1    20071115    US 2006-511718          20060728
PRAI US 2002-372450P        P     20020416
     WO 2003-CA566          W     20030416
RE.CNT  3     THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD
              ALL CITATIONS AVAILABLE IN THE RE FORMAT


=> s mycobacterium and ((alanine dehydrogenase)or(glutamine synthetase)or(serine
dehydratase))
L8          543 MYCOBACTERIUM AND ((ALANINE DEHYDROGENASE) OR(GLUTAMINE SYNTHETA
                SE) OR(SERINE DEHYDRATASE))

=> s l8 and recombinant?
L9          112 L8 AND RECOMBINANT?

=> dup rem l9
PROCESSING COMPLETED FOR L9
L10          32 DUP REM L9 (80 DUPLICATES REMOVED)

=> d bib ab kwic 1-
YOU HAVE REQUESTED DATA FROM 32 ANSWERS - CONTINUE? Y/(N):y


L10  ANSWER 1 OF 32  CAPLUS  COPYRIGHT 2009 ACS on STN
AN   2008:698876  CAPLUS <<LOGINID::20090416>>
DN   149:2678
TI   Genes and their homologs conferring trait-improving characteristics for
     plant improvement
IN   Abad, Mark; Chittoor, Jaishree; Goldman, Barry; Joseph, Mitchell; Rich,
     Ronald; Shaikh, Faten; Wray, Diana; Coffin, Marie
PA   Monsanto Technology, Llc, USA
SO   PCT Int. Appl., 162pp.
     CODEN: PIXXD2
DT   Patent
LA   English
FAN.CNT 1
     PATENT NO.          KIND   DATE        APPLICATION NO.         DATE
     ---------------     ----   --------    --------------------    --------
PI   WO 2008070179        A2    20080612    WO 2007-US25081         20071206
     WO 2008070179        A3    20081120
         W:  AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA,
             CH, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI,
             GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG,
             KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME,
             MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PG, PH, PL,
             PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, SV, SY, TJ, TM, TN,
             TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW
         RW: AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE,
             IS, IT, LT, LU, LV, MC, MT, NL, PL, PT, RO, SE, SI, SK, TR, BF,
             BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW,
             GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ,
             BY, KG, KZ, MD, RU, TJ, TM, AP, EA, EP, OA
     US 20080295196        A1    20081127    US 2007-1025            20071206
```

PRAI US 2006-873247P P 20061206

AB One hundred ninety-eight genomic DNAs or cDNAs are identified from plant, bacterial, and yeast sources that confer improved traits in Arabidopsis thaliana when expressed from sense and/or antisense constructs. Further, 19,544 homologs are identified from 1383 species. Improved traits include enhanced water use efficiency, enhanced cold or heat tolerance, enhanced reistance to salt, enhanced shade tolerance, improved yield, enhanced nitrogen use efficiency, increased seed protein or oil, enhanced herbicide resistance, and enhanced resistance to disease caused by Mol de Rio Cuarto virus or Puccinia sorghi fungus. Transgenic seed for crops with improved traits are provided by trait-improving ***recombinant*** DNA in the nucleus of cells of the seed where plants grown from such transgenic seed exhibit one or more improved traits as compared to a control plant. Of particular interest are transgenic plants that have increased yield. The present invention also provides ***recombinant*** DNA mols. for expression of a protein, and ***recombinant*** DNA mols. for suppression of a protein.

AB . . . Mol de Rio Cuarto virus or Puccinia sorghi fungus. Transgenic seed for crops with improved traits are provided by trait-improving ***recombinant*** DNA in the nucleus of cells of the seed where plants grown from such transgenic seed exhibit one or more. . . compared to a control plant. Of particular interest are transgenic plants that have increased yield. The present invention also provides ***recombinant*** DNA mols. for expression of a protein, and ***recombinant*** DNA mols. for suppression of a protein.

IT Methanococcus voltae
Methanopyrus kandleri
Methanosarcina acetivorans
Methanosarcina barkeri
Methanosarcina mazei
Methanothermobacter thermautotrophicus
Methylobacillus
Methylobacillus flagellatus
Methylobacter marinus
Methylobacterium dichloromethanicum
Methylobacterium extorquens
Methylococcus capsulatus
Microbacterium arborescens
Microbispora rosea aerata
Micrococcus luteus
Microcystis aeruginosa
Microcystis viridis
Microcystis wesenbergii
Microdochium nivale
Micromonospora echinospora
Microscilla
Mimosa pudica
Misopates orontium
Momordica charantia
Monacrosporium haptotylum
Monascus purpureus
Monilinia fructigena
Moniliophthora perniciosa
Moorella thermoacetica
Moraxella
Moricandia nitens
Moritella marina

Morus alba
Mucor mucedo
Mucor racemosus
Musa acuminata
Musa balbisiana
Musa paradisiaca
Mycobacterium
Mycobacterium abscessus
Mycobacterium avium
Mycobacterium avium paratuberculosis
Mycobacterium bovis
Mycobacterium intracellulare
Mycobacterium leprae
Mycobacterium marinum
Mycobacterium smegmatis
Mycobacterium tuberculosis
Mycoplasma arthritidis
Mycoplasma flocculare
Mycoplasma gallisepticum
Mycoplasma genitalium
Mycoplasma hominis
Mycoplasma hyopneumoniae
Mycoplasma hyorhinis
Mycoplasma mobile
Mycoplasma mycoides mycoides
Mycoplasma penetrans
Mycoplasma pirum
Mycoplasma pneumoniae
Mycoplasma pulmonis
Myxococcus xanthus
Nakaseomyces delphensis
Nannochloris bacillaris
Nanoarchaeum equitans
Narcissus pseudo-narcissus
Natrialba asiatica
Neisseria cinerea
Neisseria flavescens
Neisseria gonorrhoeae
Neisseria lactamica
Neisseria meningitidis
Neisseria mucosa
Neisseria pharyngis flava
Neisseria polysaccharea
Nelumbo nucifera
Neorickettsia sennetsu
Nepenthes alata
Nephromopsis laureri
Nephromopsis pallescens
Nephroselmis olivacea
Neurospora crassa
Neurospora terricola
Nicotiana alata
Nicotiana benthamiana
Nicotiana glauca
Nicotiana glutinosa
Nicotiana langsdorffii
Nicotiana paniculata

Nicotiana plumbaginifolia
Nicotiana sanderae
Nicotiana sylvestris
Nicotiana tabacum
Nicotiana tomentosiformis
Nitrobacter vulgaris
Nitrosomonas
Nitrosomonas europaea
Nitrosospira
Nitrosospira multiformis
Nitrosovibrio
Nitzschia
Nocardia farcinica
Nocardioides
Nodularia spumigena
Nonomuraea
Nostoc
Nostoc commune
Nostoc punctiforme
Novosphingobium aromaticivorans
Nymphaea alba
Oceanicola granulosus
Oceanobacillus iheyensis
Oceanobacter
Oceanospirillum
Ochromonas danica
Odontella sinensis
Oemleria cerasiformis
Oenococcus oeni
Oenothera elata hookeri
Ogataea minuta minuta
Olea europaea
Oligotropha carboxidovorans
Olimarabidopsis pumila
Olive
Oltmannsiellopsis viridis
Onion
Onion yellows phytoplasma
Orange
Oryza australiensis
Oryza coarctata
Oryza longistaminata
Oryza meyeriana
Oryza rufipogon
Oryza sativa
Oryza sativa indica
Oryza sativa japonica
Ostreococcus tauri
Oxyrrhis marina
Ozonium
Pachysolen tannophilus
Paenibacillus
Paenibacillus polymyxa
Panax ginseng
Pandanus amaryllifolius
Panicum maximum
Pantoea agglomerans

Pantoea dispersa
Papaver somniferum
Papaya
Paracoccidioides brasiliensis
Paracoccus denitrificans
Parmotrema perlatum
Parmotrema reticulatum
Parsley
Parvularcula bermudensis
Pasteurella multocida multocida
Pavlova lutheri
Paxillus filamentosus
Paxillus involutus
Pea
Peach
Peanut
Pear
Pectobacterium carotovorum atrosepticum
Pediococcus pentosaceus
Pelagibacter ubique
Penicillium chrysogenum
Penicillium janthinellum
Penicillium marneffei
Penicillium minioluteum
Perilla frutescens
Persea americana
Petroselinum crispum
Petunia axillaris
Petunia axillaris axillaris
Petunia hybrida
Petunia inflata
Phaeodactylum tricornutum
Phaeosphaeria avenaria triticae
Phaeosphaeria nodorum
Phaffia rhodozyma
Phanerochaete chrysosporium
Phaseolus acutifolius
Phaseolus vulgaris
Phaseolus vulgaris nanus
Philodendron oxycardium
Phleum pratense
Pholiota nameko
Phoma betae
Phoma eupyrena
Phoma herbarum
Phormidium lapideum
Photobacterium
Photobacterium leiognathi
Photobacterium phosphoreum
Photobacterium profundum
Photorhabdus luminescens
Photorhabdus luminescens laumondii
Photorhabdus temperata
Physalis crassifolia
Physalis longifolia
Physcomitrella patens
Physcomitrella patens patens

Phytophthora brassicae
Phytophthora infestans
Phytophthora nicotianae
Phytophthora palmivora
Picea abies
Picea mariana
Picea rubens
Pichia angusta
Pichia anomala
Pichia ciferrii
Pichia guilliermondii
Pichia ofunaensis
Pichia pastoris
Picrophilus torridus
Pimelobacter
Pimpinella brachycarpa
Pinus banksiana
Pinus contorta
Pinus pinaster
Pinus resinosa
Pinus strobus
Pinus sylvestris
Pinus taeda
Pinus thunbergii
Piper betel
Piromyces
Pisum sativum
Plantago major
Plantain
Platanus acerifolia
Platismatia glauca
Plectospira myriandra
Pleurotus djamor
Pleurotus eryngii
Pleurotus ostreatus
Pleurotus sajor-caju
Pneumocystis carinii
Poa pratensis
Podospora anserina
Polarella glacialis
(***genes*** and their homologs conferring trait-improving characteristics for plant improvement)

IT 9000-83-3, ATPase 9000-90-2, .alpha.-Amylase 9001-50-7, Glyceraldehyde 3-phosphate dehydrogenase 9001-52-9, Fructose 1,6-bisphosphatase 9001-59-6, Pyruvate kinase 9001-68-7 9012-90-2, DNA polymerase .alpha. 9014-01-1, Subtilisin 9023-70-5, ***Glutamine*** ***synthetase*** 9023-78-3, Triose phosphate isomerase 9023-83-0, Ribose 5-phosphate isomerase 9023-88-5 9024-52-6, Fructose bisphosphate aldolase 9026-04-4, Rhodanese 9026-51-1, Nucleoside diphosphate kinase 9028-04-0, Ubiquinone reductase 9028-83-5, D-2-Hydroxy acid dehydrogenase 9028-86-8, Aldehyde dehydrogenase 9031-50-9, Nucleotidyltransferase 9031-66-7, Aminotransferase 9031-72-5, Alcohol dehydrogenase 9032-62-6, Phosphoglycerate mutase 9033-25-4, Methyltransferase 9035-51-2, Cytochrome P 450, biological studies 9046-67-7, Serine carboxypeptidase 9059-32-9, GTPase 9075-65-4, Glycerol-3-phosphate dehydrogenase 9076-73-7, Fatty acid hydroxylase 9076-81-7, RRNA adenosine dimethyltransferase 37205-61-1, Proteinase

inhibitor 37257-07-1, .DELTA.24-Sterol methyltransferase 37259-58-8, Serine esterase 37278-25-4, RNase T2 39419-81-3, Biotin protein ligase 51845-48-8, Cyclopropane fatty acyl phospholipid synthase 56093-17-5, Ketopantoate hydroxymethyltransferase 56214-35-8, GTP cyclohydrolase II 56467-83-5, Ceramidase 78169-47-8, Aspartyl proteinase 80449-02-1, Protein tyrosine kinase 106640-75-9, Aldo/keto reductase 118390-59-3, Allene oxide cyclase 130961-00-1, 3,4-Dihydroxy-2-butanone 4-phosphate synthase 139639-26-2, Lipoate protein ligase 196717-99-4, Prenylcysteine lyase 361540-77-4, Calcineurin 372092-80-3, Protein kinase 475678-93-4, Short chain dehydrogenase
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(domain or motif; genes and their homologs conferring trait-improving characteristics for plant improvement)

L10 ANSWER 2 OF 32 SCISEARCH COPYRIGHT (c) 2009 The Thomson Corporation on STN
AN 2008:1438413 SCISEARCH <<LOGINID::20090416>>
GA The Genuine Article (R) Number: 381CU
TI Possible Involvement of an Extracellular Superoxide Dismutase (SodA) as a Radical Scavenger in Poly(cis-1,4-Isoprene) Degradation
AU Steinbuechel, Alexander (Reprint)
CS Univ Munster, Inst Mol Mikrobiol & Biotechnol, Correnstr 3, D-48149 Munster, Germany (Reprint)
E-mail: steinbu@uni-muenster.de
AU Schulte, Carina; Arenskoetter, Matthias; Berekaa, Mahmoud M.; Arenskoetter, Quyen; Priefert, Horst; Steinbuechel, Alexander (Reprint)
CS Univ Munster, Inst Mol Mikrobiol & Biotechnol, D-48149 Munster, Germany
E-mail: steinbu@uni-muenster.de
CYA Germany
SO APPLIED AND ENVIRONMENTAL MICROBIOLOGY, (DEC 2008) Vol. 74, No. 24, pp. 7643-7653.
ISSN: 0099-2240.
PB AMER SOC MICROBIOLOGY, 1752 N ST NW, WASHINGTON, DC 20036-2904 USA.
DT Article; Journal
LA English
REC Reference Count: 58
ED Entered STN: 1 Jan 2009
Last Updated on STN: 1 Jan 2009
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS
AB Gordonia westfalica Kb1 and Gordonia polyisoprenivorans VH2 induce the formation of an extracellular superoxide dismutase (SOD) during poly(cis-1,4-isoprene)degradation. To investigate the function of this enzyme in G. polyisoprenivorans VH2, the sodA gene was disrupted. The mutants exhibited reduced growth in liquid mineral salt media containing poly(cis-1,4-isoprene) as the sole carbon and energy source, and no SOD activity was detectable in the supernatants of the cultures. Growth experiments revealed that SodA activity is required for optimal growth on poly(cis-1,4-isoprene), whereas this enzyme has no effect on aerobic growth in the presence of water-soluble substrates like succinate, acetate, and propionate. This was detected by activity staining, and proof of expression was by antibody detection of SOD. When SodA from G. westfalica Kb1 was heterologously expressed in the sodA sodB double mutant Escherichia coli QC779, the ***recombinant*** mutant exhibited increased resistance to paraquat, thereby indicating the functionality of the G. westfalica Kb1 SodA and indirectly protection of G. westfalica cells by SodA from oxidative damage. Both sodA from G. polyisoprenivorans VH2 and sodA from G. westfalica Kb1 coded for polypeptides comprising 209

amino acids and having approximately 90% and 70% identical amino acids, respectively, to the SodA from ***Mycobacterium*** smegmatis strain MC2 155 and Micrococcus luteus NCTC 2665. As revealed by activity staining experiments with the wild type and the disruption mutant of G. polyisoprenivorans, this bacterium harbors only one active SOD belonging to the manganese family. The N-terminal sequences of the extracellular SodA proteins of both Gordonia species showed no evidence of leader peptides for the mature proteins, like the intracellular SodA protein of G. polyisoprenivorans VH2, which was purified under native conditions from the cells. In G. westfalica Kb1 and G. polyisoprenivorans VH2, SodA probably provides protection against reactive oxygen intermediates which occur during degradation of poly(cis-1,4-isoprene).

AB . . . SOD. When SodA from G. westfalica Kb1 was heterologously expressed in the sodA sodB double mutant Escherichia coli QC779, the ***recombinant*** mutant exhibited increased resistance to paraquat, thereby indicating the functionality of the G. westfalica Kb1 SodA and indirectly protection of. . . for polypeptides comprising 209 amino acids and having approximately 90% and 70% identical amino acids, respectively, to the SodA from ***Mycobacterium*** smegmatis strain MC2 155 and Micrococcus luteus NCTC 2665. As revealed by activity staining experiments with the wild type and. . .

STP KeyWords Plus (R): NATURAL-RUBBER LATEX; DNA-BINDING PROTEIN; SP STRAIN K30; ESCHERICHIA-COLI; ***MYCOBACTERIUM*** -TUBERCULOSIS; OXIDATIVE STRESS; ***GLUTAMINE*** - ***SYNTHETASE*** ; NOCARDIA-ASTEROIDES; GENUS GORDONIA; XANTHOMONAS SP

L10 ANSWER 3 OF 32 SCISEARCH COPYRIGHT (c) 2009 The Thomson Corporation on STN

AN 2008:1284984 SCISEARCH <<LOGINID::20090416>>

GA The Genuine Article (R) Number: 361YW

TI A Replication-Limited ***Recombinant*** ***Mycobacterium*** bovis BCG Vaccine against Tuberculosis Designed for Human Immunodeficiency Virus-Positive Persons Is Safer and More Efficacious than BCG

AU Horwitz, Marcus A. (Reprint)

CS Univ Calif Los Angeles, Sch Med, Div Infect Dis, Dept Med, CHS 37-121, 10833 Le Conte Ave, Los Angeles, CA 90095 USA (Reprint)
E-mail: MHorwitz@mednet.ucla.edu

AU Tullius, Michael V.; Harth, Guenter; Maslesa-Galic, Sasa; Dillon, Barbara J.; Horwitz, Marcus A. (Reprint)

CS Univ Calif Los Angeles, Sch Med, Div Infect Dis, Dept Med, Los Angeles, CA 90095 USA
E-mail: MHorwitz@mednet.ucla.edu

CYA USA

SO INFECTION AND IMMUNITY, (NOV 2008) Vol. 76, No. 11, pp. 5200-5214.
ISSN: 0019-9567.

PB AMER SOC MICROBIOLOGY, 1752 N ST NW, WASHINGTON, DC 20036-2904 USA.

DT Article; Journal

LA English

REC Reference Count: 53

ED Entered STN: 14 Nov 2008
Last Updated on STN: 14 Nov 2008

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Tuberculosis is the leading cause of death in AIDS patients, yet the current tuberculosis vaccine, ***Mycobacterium*** bovis bacillus Calmette-Guerin (BCG), is contraindicated for immunocompromised individuals, including human immunodeficiency virus-positive persons, because it can cause disseminated disease; moreover, its efficacy is

suboptimal. To address these problems, we have engineered BCG mutants that grow normally in vitro in the presence of a supplement, are preloadable with supplement to allow limited growth in vivo, and express the highly immunoprotective ***Mycobacterium*** tuberculosis 30-kDa major secretory protein. The limited replication in vivo renders these vaccines safer than BCG in SCID mice yet is sufficient to induce potent cell-mediated and protective immunity in the outbred guinea pig model of pulmonary tuberculosis. In the case of one vaccine, rBCG(mbtB) 30, protection was superior to that with BCG (0.3-log fewer CFU of M. tuberculosis in the lung [P < 0.04] and 0.6-log fewer CFU in the spleen [P = 0.001] in aerosol-challenged animals [means for three experiments]); hence, rBCG(mbtB) 30 is the first live mycobacterial vaccine that is both more attenuated than BCG in the SCID mouse and more potent than BCG in the guinea pig. Our study demonstrates the feasibility of developing safer and more potent vaccines against tuberculosis. The novel approach of engineering a replication-limited vaccine expressing a ***recombinant*** immunoprotective antigen and preloading it with a required nutrient, such as iron, that is capable of being stored should be generally applicable to other live vaccine vectors targeting intracellular pathogens.

TI A Replication-Limited ***Recombinant*** ***Mycobacterium*** bovis BCG Vaccine against Tuberculosis Designed for Human Immunodeficiency Virus-Positive Persons Is Safer and More Efficacious than BCG

AB Tuberculosis is the leading cause of death in AIDS patients, yet the current tuberculosis vaccine, ***Mycobacterium*** bovis bacillus Calmette-Guerin (BCG), is contraindicated for immunocompromised individuals, including human immunodeficiency virus-positive persons, because it can cause disseminated disease;. . . the presence of a supplement, are preloadable with supplement to allow limited growth in vivo, and express the highly immunoprotective ***Mycobacterium*** tuberculosis 30-kDa major secretory protein. The limited replication in vivo renders these vaccines safer than BCG in SCID mice yet. . . feasibility of developing safer and more potent vaccines against tuberculosis. The novel approach of engineering a replication-limited vaccine expressing a ***recombinant*** immunoprotective antigen and preloading it with a required nutrient, such as iron, that is capable of being stored should be. . .

STP KeyWords Plus (R): GREATER PROTECTIVE IMMUNITY; MAJOR SECRETORY PROTEIN; PANTOTHENATE AUXOTROPH; ***GLUTAMINE*** - ***SYNTHETASE*** ; GUINEA-PIGS; EXTRACELLULAR PROTEINS; MUTANT STRAIN; TB VACCINE; MODEL; RESISTANCE

L10 ANSWER 4 OF 32 BIOSIS COPYRIGHT (c) 2009 The Thomson Corporation on STN DUPLICATE 1

AN 2009:101074 BIOSIS <<LOGINID::20090416>>

DN PREV200900101074

TI Establishment of ***Glutamine*** ***Synthetase*** of ***Mycobacterium*** smegmatis as a Protein Acetyltransferase utilizing Polyphenolic Acetates as the Acetyl Group Donors.

AU Gupta, Garima; Baghel, Anil Singh; Bansal, Seema; Tyagi, Tapesh Kumar; Kumari, Ranju; Saini, Neeraj Kumar; Ponnan, Prija; Kumar, Ajit; Bose, Mridula; Saluja, Daman; Patkar, Shamkant Anant; Parmar, Virinder Singh; Raj, Hanumantharao Guru [Reprint Author]

CS Univ Delhi, Vallabhbhai Patel Chest Inst, Dept Biochem and Microbiol, Delhi 110007, India
rajhg@yahoo.com

SO Journal of Biochemistry (Tokyo), (DEC 2008) Vol. 144, No. 6, pp. 709-715. CODEN: JOBIAO. ISSN: 0021-924X.

DT Article
LA English
ED Entered STN: 4 Feb 2009
Last Updated on STN: 25 Mar 2009
AB Acetoxy Drug: Protein Transacetylase (TAase) mediating the transfer of acetyl group(s) from polyphenolic acetates (PA) to certain functional proteins in mammalian cells was identified by our earlier investigations. TAase activity was characterized in the cell lysates of ***Mycobacterium*** smegmatis and the purified protein was found to have M-r 58,000. TAase catalysed protein acetylation by a model acetoxy drug 7,8-diacetoxy-4-methylcoumarin (DAMC) was established by the demonstration of immunoreactivity of the acetylated target protein with an anti-acetyllysine antibody. The specificity of the TAase of M. smegmatis (MTAase) to various acetoxycoumarins was found to be in the order DAMC 7-AMC 6-AMC 4-AC 3-AC ABP. Also, the N-terminal sequence of purified MTAase was found to perfectly match with ***glutamine*** ***synthetase*** (GS) of M. smegmatis. The identity of MTAase with GS was confirmed by the observation that the purified MTAase as well as the purified ***recombinant*** GS exhibited all the properties of GS. The finding that purified Escherichia coli GS was found to have substantial TAase activity highlighted the TAase function of GS in other bacteria. These results conclusively established for the first time the protein acetyltransferase function of GS of M. smegmatis.
TI Establishment of ***Glutamine*** ***Synthetase*** of ***Mycobacterium*** smegmatis as a Protein Acetyltransferase utilizing Polyphenolic Acetates as the Acetyl Group Donors.
AB. . . functional proteins in mammalian cells was identified by our earlier investigations. TAase activity was characterized in the cell lysates of ***Mycobacterium*** smegmatis and the purified protein was found to have M-r 58,000. TAase catalysed protein acetylation by a model acetoxy drug. . . order DAMC 7-AMC 6-AMC 4-AC 3-AC ABP. Also, the N-terminal sequence of purified MTAase was found to perfectly match with ***glutamine*** ***synthetase*** (GS) of M. smegmatis. The identity of MTAase with GS was confirmed by the observation that the purified MTAase as well as the purified ***recombinant*** GS exhibited all the properties of GS. The finding that purified Escherichia coli GS was found to have substantial TAase. . .
IT Major Concepts
Pharmacology; Enzymology (Biochemistry and Molecular Biophysics)
IT Chemicals & Biochemicals
glutamine ***synthetase*** [EC 6.3.1.2]; polyphenolic acetate; acetyl group donor; protein acetyltransferase: enzyme inhibitor-drug; 7,8-diacetoxy-4-methylcoumarin: enzyme inhibitor-drug
ORGN . . .
Notes
Bacteria, Eubacteria, Microorganisms
ORGN Classifier
Mycobacteriaceae 08881
Super Taxa
Mycobacteria; Actinomycetes and Related Organisms; Eubacteria; Bacteria; Microorganisms
Organism Name
Mycobacterium smegmatis (species): strain-VT-301
Taxa Notes
Bacteria, Eubacteria, Microorganisms

RN 9023-70-5 (***glutamine*** ***synthetase***)
9023-70-5 (EC 6.3.1.2)
116155-74-9 (protein acetyltransferase)

L10 ANSWER 5 OF 32 BIOSIS COPYRIGHT (c) 2009 The Thomson Corporation on STN
DUPLICATE 2
AN 2008:409086 BIOSIS <<LOGINID::20090416>>
DN PREV200800409085
TI Overexpression, purification, crystallization and preliminary X-ray analysis of Rv2780 from ***Mycobacterium*** tuberculosis H37Rv.
AU Tripathi, Sarvind Mani; Ramachandran, Ravishankar [Reprint Author]
CS Cent Drug Res Inst, Mol and Struct Biol Div, POB 173,Chattar Manzil,Mahatma Gandhi Marg, Lucknow 226001, Uttar Pradesh, India
r_ravishankar@cdri.res.in
SO Acta Crystallographica Section F Structural Biology and Crystallization Communications, (MAY 2008) Vol. 64, No. Part 5, pp. 367-370.
ISSN: 1744-3091. E-ISSN: 1744-3091.
DT Article
LA English
ED Entered STN: 31 Jul 2008
Last Updated on STN: 31 Jul 2008
AB Rv2780, an ***alanine*** ***dehydrogenase*** from ***Mycobacterium*** tuberculosis (MtAlaDH), catalyzes the NAD-dependent interconversion of alanine and pyruvate. ***Alanine*** ***dehydrogenase*** is released into the culture medium in substantial amounts by virulent strains of mycobacteria and is not found in the vaccine strain of tuberculosis. Crystals of ***recombinant*** MtAlaDH were grown from 2 M ammonium sulfate solution at similar to 12 mg ml(-1) protein concentration in two crystal forms which occur in the presence and absence of NAD/pyruvate, respectively. Diffraction data extending to 2.6 angstrom were collected at room temperature from both apo and ternary complex crystals. Crystals of the apoenzyme have unit-cell parameters a = 173.89, b = 127.07, c = 135.95 angstrom. They are rod-like in shape and belong to space group C2. They contain a hexamer in the asymmetric unit. Crystals of the ternary complex belong to space group P4(3)2(1)2 and have unit-cell parameters a = b = 88.99, c = 373.85 angstrom. There are three subunits in the asymmetric unit of the holoenzyme crystals.
TI Overexpression, purification, crystallization and preliminary X-ray analysis of Rv2780 from ***Mycobacterium*** tuberculosis H37Rv.
AB Rv2780, an ***alanine*** ***dehydrogenase*** from ***Mycobacterium*** tuberculosis (MtAlaDH), catalyzes the NAD-dependent interconversion of alanine and pyruvate. ***Alanine*** ***dehydrogenase*** is released into the culture medium in substantial amounts by virulent strains of mycobacteria and is not found in the vaccine strain of tuberculosis. Crystals of ***recombinant*** MtAlaDH were grown from 2 M ammonium sulfate solution at similar to 12 mg ml(-1) protein concentration in two crystal. . .
IT . . . Concepts
Methods and Techniques; Molecular Genetics (Biochemistry and Molecular Biophysics); Enzymology (Biochemistry and Molecular Biophysics)
IT Chemicals & Biochemicals
Rv2780: ***alanine*** ***dehydrogenase*** , expression, crystallization, purification
ORGN Classifier
Mycobacteriaceae 08881

Super Taxa
Mycobacteria; Actinomycetes and Related Organisms; Eubacteria; Bacteria; Microorganisms
Organism Name
Mycobacterium tuberculosis H37Rv (species)
Taxa Notes
Bacteria, Eubacteria, Microorganisms

GEN ***Mycobacterium*** tuberculosis H37Rv ald gene [***Mycobacterium*** tuberculosis H37Rv Rv2780 gene] (Mycobacteriaceae): expression

L10 ANSWER 6 OF 32 CAPLUS COPYRIGHT 2009 ACS on STN
AN 2007:593435 CAPLUS <<LOGINID::20090416>>
DN 146:516103
TI Polynucleotides and polypeptides useful for improved agronomic traits in transgenic plants
IN Abad, Mark Scott; Chelf, Frances; Coffin, Marie A.; Darveaux, Bettina; Goldman, Barry S.; McDonald, Maria; Rich, Ronald; Slaten, Erin; Wilkins, Shanita
PA USA
SO U.S. Pat. Appl. Publ., 81pp.
CODEN: USXXCO
DT Patent
LA English
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 20070124833	A1	20070531	US 2006-431855	20060510
WO 2006138005	A3	20090129	WO 2006-US18535	20060510

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, LY, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NG, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SM, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW
RW: AP, BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, EA, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, EP, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IS, IT, LT, LU, LV, MC, NL, PL, PT, RO, SE, SI, SK, TR, OA, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

PRAI US 2005-679917P	P	20050510	
US 2005-723596P	P	20051004	

AB Transgenic seed for crops with improved traits are provided by trait-improving ***recombinant*** DNA in the nucleus of cells of the seed where plants grown from such transgenic seed exhibit one or more improved traits as compared to a control plant. To identify ***recombinant*** DNA that confers improved traits to plants, Arabidopsis thaliana was transformed with a candidate ***recombinant*** DNA construct and screened for an improved trait. Desirable agronomic traits include improved water use efficiency, cold tolerance, increased yield, improved nitrogen use efficiency, increased seed protein and oil content, heat tolerance, salt resistance, shade tolerance, herbicide resistance, and resistance to viral or fungal infections. Of particular interest are transgenic plants that have increased yield. Four hundred twenty-five ***recombinant*** nucleic acids and gene products were identified. BLAST searching identified 32,784 homologs to the 425 proteins. The present invention also provides ***recombinant*** DNA

mols. for expression of a protein, and ***recombinant*** DNA mols. for suppression of a protein.

AB Transgenic seed for crops with improved traits are provided by trait-improving ***recombinant*** DNA in the nucleus of cells of the seed where plants grown from such transgenic seed exhibit one or more improved traits as compared to a control plant. To identify ***recombinant*** DNA that confers improved traits to plants, Arabidopsis thaliana was transformed with a candidate ***recombinant*** DNA construct and screened for an improved trait. Desirable agronomic traits include improved water use efficiency, cold tolerance, increased yield,. . . and resistance to viral or fungal infections. Of particular interest are transgenic plants that have increased yield. Four hundred twenty-five ***recombinant*** nucleic acids and gene products were identified. BLAST searching identified 32,784 homologs to the 425 proteins. The present invention also provides ***recombinant*** DNA mols. for expression of a protein, and ***recombinant*** DNA mols. for suppression of a protein.

IT Carica papaya
Carmichaelia
Carpinus caroliniana
Carrot
Casearia sylvestris
Cassava
Cassia fistula
Cassinopsis ilicifolia
Castanea sativa
Castanopsis inermis
Castanospermum australe
Casuarina cunninghamiana
Catharanthus roseus
Cathaya argyrophylla
Cattleya bicolor
Cattleya intermedia
Caucanthus auriculatus
Caulobacter crescentus
Caulobacter vibrioides
Caulophyllum robustum
Caulophyllum thalictroides
Cedrela odorata
Cedrus atlantica
Cedrus deodara
Celery
Cenarchaeum symbiosum
Centaurea calcitrapa
Centaurium umbellatum
Cephalomanes thysanostomum
Cephalomappa malloticarpa
Cephalopentandra ecirrhosa
Cercocarpus ledifolius
Cercospora zeae-maydis
Chadsia versicolor
Chaetosphaeridium globosum
Chamaebatiaria millefolium
Chara corallina
Cheiranthus cheiri
Chenopodium murale
Chenopodium rubrum

Chickpea
Chicory
Chimonanthus praecox
Chinese cabbage
Chlamydia muridarum
Chlamydia trachomatis
Chlamydomonas
Chlamydomonas incerta
Chlamydomonas reinhardtii
Chlamydophila caviae
Chlamydophila pneumoniae
Chloranthus nervosus
Chlorella vulgaris
Chlorobaculum tepidum
Chlorobium limicola
Chloroflexus aurantiacus
Chondrostereum purpureum
Chondrus crispus
Chorispora bungeana
Choristylis rhamnoides
Chromobacterium violaceum
Chromohalobacter salexigens
Chromolaena
Chrysanthemum lavandulaefolium
Chrysanthemum maximum
Chrysanthemum morifolium
Chrysophyllum oliviforme
Cicer arietinum
Cicer pinnatifidum
Cichorium endivia
Cichorium intybus
Cinnamomum camphora
Circaea alpina
Circaea cordata
Cirsium texanum
Citrobacter amalonaticus
Citrobacter braakii
Citrobacter freundii
Citrobacter koseri
Citrofortunella mitis
Citrullus lanatus
Citrus (genus)
Citrus aurantium
Citrus hystrix
Citrus limon
Citrus paradisi
Citrus reticulata
Citrus sinensis
Cladosporium fulvum
Cladosporium herbarum
Cladrastis sikokiana
Cladrastis sinensis
Clarkia amoena
Clarkia delicata
Clarkia dudleyana
Clarkia epilobioides
Clarkia gracilis

Clarkia heterandra
Clarkia lassenensis
Clarkia lewisii
Clarkia lingulata
Clarkia modesta
Clarkia similis
Clarkia unguiculata
Clarkia xantiana
Clavibacter michiganensis michiganensis
Claviceps fusiformis
Claviceps purpurea
Clavija eggersiana
Clavispora lusitaniae
Clethra alnifolia
Clethra barbinervis
Cliftonia monophylla
Clostridium acetobutylicum
Clostridium beijerinckii
Clostridium bifermentans
Clostridium butyricum
Clostridium cadaveris
Clostridium cellulovorans
Clostridium clostridioforme
Clostridium difficile
Clostridium histolyticum
Clostridium innocuum
Clostridium kluyveri
Clostridium longisporum
Clostridium perfringens
Clostridium ramosum
Clostridium saccharobutylicum
Clostridium saccharoperbutylacetonicum
Clostridium septicum
Clostridium sordellii
Clostridium sporogenes
Clostridium sticklandii
Clostridium tertium
Clostridium tetani
Clostridium thermocellum
Clover phyllody phytoplasma
Clusia minor
Coccidioides posadasii
Coccinia adoensis
Cocculus trilobus
Cochliobolus carbonum
Cochliobolus heterostrophus
Coconut
Cocos nucifera
Codonopsis lanceolata
Coffea arabica
Coffea canephora
Coleochaete orbicularis
Colletotrichum gloeosporioides malvae
Colletotrichum trifolii
Colwellia maris
Comamonas
Comamonas acidovorans

Comamonas testosteroni
Combretocarpus rotundatus
Comptonia peregrina
Connarus conchocarpus
Convallaria majalis
Convolvulus sepium
Coptis japonica
Corchorus capsularis
Cordyceps bassiana
Coriaria arborea
Coriaria myrtifolia
Coriaria ruscifolia
Coriaria sarmentosa
Coris monspeliensis
Corn
Cornus florida
Cornus mas
Cornus nuttallii
Cornus walteri
Corokia cotoneaster
Cortusa
Cortusa matthioli
Corydalis nobilis
Corylopsis pauciflora
Corylus avellana
Corynebacterium ammoniagenes
Corynebacterium crenatum
Corynebacterium diphtheriae
Corynebacterium efficiens
Corynebacterium glutamicum
Corynocarpus cribbianus
Corynocarpus dissimilis
Corynocarpus laevigatus
Corynocarpus similis
Corypha taliera
Corypha umbraculifera
Cotinus coggygria
Cowpea
Coxiella burnetii
Craibella phuyensis
Crambe cordifolia
Cranocarpus martii
Crataegus columbiana
Crataegus monogyna
Craterosiphon scandens
Craterostigma plantagineum
Cratoneuron filicinum
Cremastosperma microcarpum
Crenarchaeota
Crepidomanes birmanicum
Crepidomanes latealatum
Crinodendron patagua
Crocosphaera watsonii
Crocus sativus
Crossosoma californicum
Crossostylis biflora
Crucihimalaya wallichii

Cryphonectria parasitica
Cryptococcus curvatus
Cryptococcus neoformans grubii
Cryptococcus neoformans neoformans
Cryptomeria japonica
Ctenolophon englerianus
Cucumber
Cucumis anguria
Cucumis melo
Cucumis sativus
Cucurbita argyrosperma
Cucurbita argyrosperma sororia
Cucurbita digitata
Cucurbita maxima
Cucurbita moschata
Cucurbita pepo
Cunninghamella elegans
Cupriavidus metallidurans
Cupriavidus necator
Cupriavidus oxalaticus
Curtisia dentata
Cuscuta reflexa
Cussonia spicata
Cuttsia viburnea
Cyanidioschyzon merolae
Cyanidium caldarium
Cyanophora paradoxa
Cycas circinalis
Cycas revoluta
Cyclamen hederifolium
Cycloclasticus oligotrophus
Cydonia oblonga
(polynucleotides and polypeptides useful for improved agronomic traits in transgenic plants)
IT Cydonia speciosa
Cylicomorpha parviflora
Cylindrotheca fusiformis
Cymbidium
Cynara cardunculus
Cynodon dactylon
Cytophaga
Cytophaga hutchinsonii
DNA sequences
Dactylis glomerata
Dais cotinifolia
Dalbergia hupeana
Dalbergiella welwitschii
Daphne mezereum
Daphniphyllum
Darmera peltata
Dasyphyllum argenteum
Dasyphyllum dicanthoides
Datura ferox
Datura metel
Datura stramonium
Daucus carota
Davallia epiphylla

Davallia solida
Davidia involucrata
Debaryomyces hansenii
Debaryomyces occidentalis
Dechloromonas aromatica
Decumaria barbara
Decumaria sinensis
Degeneria vitiensis
Deinococcus proteolyticus
Deinococcus radiodurans
Delftia tsuruhatensis
Delphinium grandiflorum
Dendrobium
Dendrobium crumenatum
Deschampsia antarctica
Desfontainia spinosa
Desmopsis microcarpa
Desmopsis schippii
Desulfitobacterium hafniense
Desulfovibrio desulfuricans
Desulfovibrio gigas
Desulfovibrio vulgaris
Desulfovibrio vulgaris vulgaris
Deutzia gracilis
Deutzia rubens
Dewevrea bilabiata
Dianthus caryophyllus
Dianthus gratianopolitanus
Dianthus plumarius
Diapensia lapponica
Diaporthe helianthi
Dicentra eximia
Dickeya chrysanthemi
Dicranodontium denudatum
Diervilla sessilifolia
Dimorphotheca pluvialis
Dinemagonum gayanum
Dinemandra ericoides
Dionysia microphylla
Dionysia tapetodes
Dioscorea communis
Dioscorea elephantipes
Dioscorea gracillima
Dioscorea nipponica
Dioscorea quinqueloba
Dioscorea septemloba
Dioscorea tenuipes
Diospyros kaki
Diospyros virginiana
Diphylleia cymosa
Diplocyclos palmatus
Diplopeltis huegelii
Dipsacus mitis
Dipteryx odorata
Discaria chacaye
Discaria toumatou
Dodecatheon meadia

Dombeya
Doniophyton anomalum
Donnellsmithia cordata
Doritaenopsis
Dorstenia psilurus
Douglasia nivalis
Dovea macrocarpa
Dovyalis rhamnoides
Dozya japonica
Dracunculus vulgaris
Drimys winteri
Drummondia obtusifolia
Dryas drummondii
Dryopteris caudipinna
Dunaliella salina
Dunaliella tertiolecta
Dussia tessmannii
Dysosma versipellis
Ecballium elaterium
Echinochloa crus-galli formosensis
Echinochloa phyllopogon
Edgeworthia papyrifera
Eggplant
Ehrlichia canis
Ehrlichia ruminantium
Elaeagnus angustifolia
Elaeis guineensis
Elegia asperiflora
Elmera racemosa
Elodea densa
Elymus cinereus
Elymus elongatum
Elymus triticoides
Embryophyta
Emericella nidulans
Emmenosperma alphitonioides
Emorya suaveolens
Endive
Endospermum moluccanum
Enterobacter aerogenes
Enterobacter cloacae
Enterobacter gergoviae
Enterococcus casseliflavus
Enterococcus faecalis
Enterococcus faecium
Enterococcus hirae
Entodon luridus
Entodon rubicundus
Ephedra tweediana
Ephemerum spinulosum
Epifagus virginianus
Epilobium brachycarpum
Epilobium canum
Equisetum arvense
Eragrostis japonica
Eremocharis fruticosa
Eremopyrum bonaepartis

Eremopyrum distans
Eremosyne pectinata
Eremothamnus marlothianus
Eremothecium gossypii
Eriobotrya japonica
Eriocnema fulva
Erwinia
Erwinia amylovora
Erwinia pyrifoliae
Erythrophleum ivorense
Erythroxylum confusum
Escallonia coquimbensis
Escallonia pulverulenta
Escherichia albertii
Escherichia coli
Escherichia fergusonii
Escherichia vulneris
Eschscholzia californica
Eubacteria
Eucalyptus botryoides
Eucalyptus camaldulensis
Eucalyptus cordata
Eucalyptus globulus
Eucalyptus gunnii
Eucalyptus saligna
Eucommia ulmoides
Eugeissona tristis
Eugenia uniflora
Euglena gracilis
Euglena longa
Euonymus alata
Eupatorium atrorubens
Euphorbia esula
Euphorbia lagascae
Eustoma grandiflorum
Eutreptia viridis
Exbucklandia populnea
Excoecaria cochinchinensis
Exiguobacterium
Exochorda giraldii
Exophiala dermatitidis
Fagonia cretica
Fagonia indica
Fagonia luntii
Fagopyrum
Fagopyrum cymosum
Fagopyrum esculentum
Fagopyrum tataricum potanini
Fagus crenata
Fagus gunnii
Fagus sylvatica
Fallopia japonica
Fallugia paradoxa
Felicia bergeriana
Fendlera rupicola
Fendlerella utahensis
Ferroplasma acidarmanus

Fervidobacterium
Ficus carica
Fig
Filarum manserichense
Filipendula purpurea
Filipendula vulgaris
Filobasidiella neoformans
Fischerella
Flabellariopsis acuminata
Flacourtia jangomas
Flaveria bidentis
Flaveria chloraefolia
Flaveria palmeri
Flaveria pringlei
Flaveria ramosissima
Flaveria trinervia
Flavobacterium
Flavobacterium columnare
Flavobacterium johnsoniae
Fluoribacter bozemanae
Fluoribacter dumoffii
Fluoribacter gormanii
Fontinalis antipyretica
Forsythia intermedia
Fortunella margarita
Fouquieria columnaris
Fragaria ananassa
Fragaria grandiflora
Fragaria vesca
Francisella novicida
Francisella tularensis
Francisella tularensis holarctica
Francisella tularensis mediasiatica
Francisella tularensis tularensis
Frankia
Frankia alni
Frateuria
Fraxinus excelsior
Fremontodendron mexicanum
Fritillaria agrestis
Fritillaria liliacea
Fritschea bemisiae
Fuchsia cylindracea
Fuchsia cyrtandroides
Fucus distichus
Fusarium chlamydosporum
Fusarium graminearum
Fusarium lycopersici
Fusarium oxysporum
(***polynucleotides*** and polypeptides useful for improved agronomic traits in transgenic plants)

IT 9000-91-3, .beta.-Amylase 9000-96-8, Arginase 9001-16-5, Cytochrome oxidase 9001-22-3, .beta.-Glucosidase 9001-41-6, Glucose-6-phosphate isomerase 9001-47-2, Glutaminase 9001-59-6, Pyruvate kinase 9001-81-4, Phosphoglucomutase 9001-83-6, Phosphoglycerate kinase 9013-02-9, Adenylate kinase 9013-66-5, Glutathione peroxidase 9014-08-8, Enolase 9014-24-8, RNA polymerase 9014-52-2, Tryptophan

synthase 9016-12-0, Hypoxanthine phosphoribosyltransferase 9016-18-6, Carboxylesterase 9023-03-4, NADPH-Ferrihemoprotein reductase 9023-09-0, Sulfotransferase 9023-70-5, ***Glutamine*** ***synthetase*** 9023-78-3, Triose phosphate isomerase 9023-88-5, Phosphomannose isomerase 9024-20-8, Ribulose 5-phosphate-3-epimerase 9024-52-6, Fructose bisphosphate aldolase 9025-72-3, Trehalose-6-phosphate phosphatase 9027-23-0, Ribulose bisphosphate carboxylase 9027-96-7, Citrate synthase 9028-37-9, Glycerate dehydrogenase 9028-84-6, Formaldehyde dehydrogenase 9028-85-7, EC 1.2.1.2 9028-86-8, NAD-aldehyde dehydrogenase 9028-90-4, Betaine aldehyde dehydrogenase 9028-93-7, IMP dehydrogenase 9028-95-9, Succinate-semialdehyde dehydrogenase 9029-02-1, L-Galactono-1,4-lactone dehydrogenase 9029-26-9, Monodehydroascorbate reductase 9030-26-6, Nicotinate phosphoribosyltransferase 9030-40-4, Acetylornithine aminotransferase 9030-42-6 9030-45-9, Glutamine-fructose-6-phosphate aminotransferase 9030-51-7, Fructokinase 9030-70-0, Cystathionine .gamma.-synthase 9031-72-5, Alcohol dehydrogenase 9032-03-5, 5-Aminoimidazole-4-carboxamide ribonucleotide formyltransferase 9032-20-6, NADPH quinone oxidoreductase 9032-22-8 9032-62-6, Phosphoglycerate mutase 9033-12-9, Lactoylglutathione lyase 9035-51-2, Cytochrome P 450, biological studies 9037-67-6, 4-Aminobutyrate aminotransferase 9038-14-6 9044-88-6, Prephenate dehydratase 9054-82-4, 1-Pyrroline-5-carboxylate dehydrogenase 9055-30-5, 2-Phospho-D-glycerate hydrolyase 9055-46-3, Dihydrodipicolinate reductase 9073-94-3, Phosphoenolpyruvate carboxykinase 9073-95-4, Phosphogluconate dehydrogenase 9075-68-7, Pullulanase 9076-57-7 37213-53-9 37255-37-1, .DELTA.7-Sterol C5-desaturase 37255-38-2, Glutaryl-CoA dehydrogenase 37256-51-2, Sulfite reductase 37259-80-6, Demethylmenaquinone methyltransferase 37278-24-3, GDP-mannose pyrophosphorylase 37289-22-8, EC 3.5.4.19 37290-89-4, Cysteine synthase 37353-36-9, Acetyl-coenzyme A:acetoacetyl-coenzyme A transferase 39279-34-0 55467-36-2, Cinnamyl alcohol dehydrogenase 56467-83-5, Ceramidase 63551-76-8, Phosphoinositide phospholipase C 67880-93-7, Mercuric reductase 68518-07-0, Glutamate 1-semialdehyde 2,1-aminomutase 78310-66-4, NADPH-methylglyoxal reductase 84012-74-8, D-Cysteine desulfhydrase 85638-48-8, Diadenosine tetraphosphate hydrolase 86280-59-3, Glycerophosphoryl diester phosphodiesterase 86480-67-3, Ubiquitin C-terminal hydrolase 88414-92-0, .beta.-Ketoacyl-CoA synthase 95076-93-0, Peptidylprolyl cis-trans isomerase 101150-03-2, 12-Oxophytodienoate reductase 109136-49-4, Ubiquitin-specific protease 109301-01-1, Glyoxal oxidase 122097-10-3, Ferric-chelate reductase 134549-83-0 140879-24-9, Proteasome 187042-30-4, Calcium-dependent protein kinase 192230-91-4, Protein kinase MPK4 197462-59-2, Myrcene synthase 209864-08-4, L-Galactose dehydrogenase 228273-07-2, HAL5 protein kinase 362674-81-5 362690-38-8, Protein phosphatase 2C 366806-33-9, Casein kinase II 378782-09-3, Cytochrome P 450 98A3 414863-56-2, Protein O-fucosyltransferase 1 475678-93-4, Short-chain dehydrogenase/reductase 929259-81-4
RL: AGR (Agricultural use); BIOL (Biological study); USES (Uses)
(polynucleotides and polypeptides useful for improved agronomic traits in transgenic plants)

L10 ANSWER 7 OF 32 CAPLUS COPYRIGHT 2009 ACS on STN
AN 2006:1311631 CAPLUS <<LOGINID::20090416>>
DN 146:6454
TI Manufacture of L-amino acids with ***recombinant*** microorganism by

enzymic resolution
IN Hayashi, Motoko; Yamamoto, Hiroaki; Kimoto, Norihiro
PA Daicel Chemical Industries, Ltd., Japan
SO PCT Int. Appl., 98pp.
CODEN: PIXXD2
DT Patent
LA Japanese
FAN.CNT 1

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	WO 2006132145	A1	20061214	WO 2006-JP311081	20060602
	W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, LY, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NG, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SM, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW				
	RW: AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IS, IT, LT, LU, LV, MC, NL, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
	EP 1900821	A1	20080319	EP 2006-756916	20060602
	R: DE				
	CN 101194020	A	20080604	CN 2006-80020215	20071207
PRAI	JP 2005-169919	A	20050609		
	WO 2006-JP311081	W	20060602		

AB L-amino acids are manufd. from D-amino acids by first conversion to keto-acids and then to L-amino acids with ***recombinant*** microorganism such as Escherichia coli harboring D-amino acid oxidase and/or D-amino acid dehydrogenase, and L-amino acid dehydrogenase and/or L-amino acid aminotransferase. Also the ***recombinant*** microorganism harboring gene(S) for regeneration of coenzyme such as formate dehydrogenase and H2O2-degrading enzyme such as catalase. Construction of plasmids contg. genes and prepn. of ***recombinant*** E. coli for prepn. of L-norvaline from D-norvaline or DL-norvaline were shown.
RE.CNT 5 THERE ARE 5 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT
TI Manufacture of L-amino acids with ***recombinant*** microorganism by enzymic resolution
AB L-amino acids are manufd. from D-amino acids by first conversion to keto-acids and then to L-amino acids with ***recombinant*** microorganism such as Escherichia coli harboring D-amino acid oxidase and/or D-amino acid dehydrogenase, and L-amino acid dehydrogenase and/or L-amino acid aminotransferase. Also the ***recombinant*** microorganism harboring gene(S) for regeneration of coenzyme such as formate dehydrogenase and H2O2-degrading enzyme such as catalase. Construction of plasmids contg. genes and prepn. of ***recombinant*** E. coli for prepn. of L-norvaline from D-norvaline or DL-norvaline were shown.
ST amino acid enzymic resoln ***recombinant*** microorganism
IT Bacillus thermocellulolyticus
Candida boidinii
Escherichia coli
Fermentation

Geobacillus stearothermophilus
Lysinibacillus sphaericus
Mycobacterium vaccae
Shewanella
Thermoactinomyces intermedius
pH
(manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)

IT Amino acids, preparation
RL: BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation)
(manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)

IT Gene, microbial
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)

IT Carboxylic acids, biological studies
RL: BPN (Biosynthetic preparation); BSU (Biological study, unclassified); RCT (Reactant); BIOL (Biological study); PREP (Preparation); RACT (Reactant or reagent)
(oxo; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)

IT Plasmids
(pETECDD1; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)

IT Plasmids
(pFGSLED1; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)

IT Plasmids
(pSE-BSB1; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)

IT Plasmids
(pSE-ECB1; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)

IT Plasmids
(pSE420Q; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)

IT Plasmids
(pSF-BTA-1; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)

IT Plasmids
(pSF-GSA2; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)

IT Plasmids
(pSF-SAD1; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)

IT Plasmids
(pSF-TIP2; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)

IT Plasmids
(pSFBPAD1; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)

IT Plasmids
(pSFBPLD1; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)

IT Plasmids

(pSFCPC01; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)
IT Plasmids
(pSFGACO1; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)
IT Plasmids
(pSFTPCO1; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)
IT Plasmids
(pSQECKE1; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)
IT Plasmids
(pSQECKG1; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)
IT Plasmids
(pSU-MF26; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)
IT Plasmids
(pSUCBDO1; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)
IT Plasmids
(pSUTVDO1; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)
IT Amino acids, biological studies
RL: BSU (Biological study, unclassified); RCT (Reactant); BIOL (Biological study); RACT (Reactant or reagent)
(D-; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)
IT Amino acids, biological studies
RL: BSU (Biological study, unclassified); RCT (Reactant); BIOL (Biological study); RACT (Reactant or reagent)
(DL-amino acids; manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)
IT 6600-40-4P, L-Norvaline
RL: BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation)
(manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)
IT 1821-02-9P, 2-Oxopentanoic acid
RL: BPN (Biosynthetic preparation); BSU (Biological study, unclassified); RCT (Reactant); BIOL (Biological study); PREP (Preparation); RACT (Reactant or reagent)
(manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)
IT 7722-84-1, Hydrogen peroxide, biological studies
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)
IT 760-78-1, Norvaline 2013-12-9, D-Norvaline
RL: BSU (Biological study, unclassified); RCT (Reactant); BIOL (Biological study); RACT (Reactant or reagent)
(manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)
IT 9000-88-8, D-Amino acid oxidase 9001-05-2, Catalase 9028-85-7, Formate dehydrogenase 9029-06-5, L- ***Alanine*** ***dehydrogenase*** 9029-13-4, L-Amino acid dehydrogenase 9031-66-7, Aminotransferase 9082-71-7, L-Leucine dehydrogenase 37205-44-0, D-Amino acid

dehydrogenase 69403-12-9, Phenylalanine dehydrogenase
RL: CAT (Catalyst use); USES (Uses)
(manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)
IT 540-69-2, Ammonium formate
RL: RCT (Reactant); RACT (Reactant or reagent)
(manuf. of L-amino acids with ***recombinant*** microorganism by enzymic resoln.)

L10 ANSWER 8 OF 32 EMBASE COPYRIGHT (c) 2009 Elsevier B.V. All rights reserved on STN
AN 2006589857 EMBASE <<LOGINID::20090416>>
TI Development of a simple high-throughput screening protocol based on biosynthetic activity of ***Mycobacterium*** tuberculosis ***glutamine*** ***synthetase*** for the identification of novel inhibitors.
AU Singh, Upasana; Sarkar, Dhiman, Dr. (correspondence)
CS Combi Chem-Bio Resource Center, National Chemical Laboratory, Dr. Homi Bhabha Rd., Pune 411008, India. d.sarkar@ncl.res.in
SO Journal of Biomolecular Screening, (Dec 2006) Vol. 11, No. 8, pp. 1035-1042.
Refs: 28
ISSN: 1087-0571 E-ISSN: 1552-454X CODEN: JBISF3
CY United States
DT Journal; Article
FS 027 Biophysics, Bioengineering and Medical Instrumentation
004 Microbiology: Bacteriology, Mycology, Parasitology and Virology
LA English
SL English
ED Entered STN: 2 Jan 2007
Last Updated on STN: 2 Jan 2007
AB A high-throughput screening protocol has been developed for Mycobactenum tuberculosis ***glutamine*** ***synthetase*** by quantitative estimation of inorganic phosphate. The K(m) values determined at pH 6.8 are 22 mM for L-glutamic acid, 0.75 mM for NH(4)Cl, 3.25 mM for MgCl(2), and 2.5 mM for adenosine triphosphate. The K(m) value for glutamine is affected significantly by the increase in pH of assay buffer. At the saturating level of the substrate, the enzyme activity at pH 6.8 and 25.degree.C is found to be linear up to 3 h. The reduction of enzyme activity is negligible even in presence of 10% DMSO. The Z' factor and signal-to-noise ratio are found to be 0.75 and 6.18, respectively, when the enzyme is used at 62.5 .mu.g/ml concentration. The IC(50) values obtained at pH 6.8 for both L-methionine S-sulfoximine and DL-phosphothriacin are 500 .mu.M and 30 .mu.M, respectively, which is lowest compared to the values obtained at other pH levels. The Beckman Coulter high-throughput screening platform was found to take 5 h 9 min to complete the screening of 60 plates. For each assay plate, a replica plate is used to normalize the data. Screening of 1164 natural product fractions/extracts and synthetic molecules from an in-house library was able to identify 12 samples as confirmed hits. Altogether, the validation data from screening of a small set of an in-house library coupled with Z' and signalto-noise values indicate that the protocol is robust for high-throughput screening of a diverse chemical library. .COPYRGT. 2006 Society for Biomolecular Sciences.
TI Development of a simple high-throughput screening protocol based on biosynthetic activity of ***Mycobacterium*** tuberculosis ***glutamine*** ***synthetase*** for the identification of novel

inhibitors.

AB A high-throughput screening protocol has been developed for Mycobactenum tuberculosis ***glutamine*** ***synthetase*** by quantitative estimation of inorganic phosphate. The K(m) values determined at pH 6.8 are 22 mM for L-glutamic acid, 0.75. . .

CT Medical Descriptors:
article
controlled study
enzyme activity
enzyme analysis
*enzyme inhibition
enzyme kinetics
enzyme regulation
enzyme substrate
*enzyme synthesis
*high throughput screening
IC 50
Michaelis constant
****Mycobacterium tuberculosis***
nonhuman
pH
priority journal
process development
quantitative assay
screening test
signal noise ratio
validation process
adenosine triphosphate
ammonium chloride
bacterial enzyme
dimethyl sulfoxide
*enzyme inhibitor
*glutamate ammonia ligase
glutamic acid
glutamine
magnesium chloride
methionine sulfoximine
natural product
recombinant enzyme

L10 ANSWER 9 OF 32 BIOSIS COPYRIGHT (c) 2009 The Thomson Corporation on STN
DUPLICATE 3

AN 2005:439093 BIOSIS <<LOGINID::20090416>>

DN PREV200510229560

TI Structure of ***Mycobacterium*** tuberculosis ***glutamine*** ***synthetase*** in complex with a transition-state mimic provides functional insights.

AU Krajewski, Wojciech W.; Jones, T. Alwyn; Mowbray, Sherry L. [Reprint Author]

CS Swedish Univ Agr Sci, Ctr Biomed, Dept Biol Mol, Box 590, SE-75124 Uppsala, Sweden
mowbray@xray.bmc.uu.se

SO Proceedings of the National Academy of Sciences of the United States of America, (JUL 26 2005) Vol. 102, No. 30, pp. 10499-10504.
CODEN: PNASA6. ISSN: 0027-8424.

DT Article

LA English

ED Entered STN: 26 Oct 2005
Last Updated on STN: 26 Oct 2005
AB ***Glutamine*** ***synthetase*** catalyzes the ligation of glutamate and ammonia to form glutamine, with the resulting hydrolysis of ATP. The enzyme is a central component of bacterial nitrogen metabolism and is a potential drug target. Here, we report a high-yield ***recombinant*** expression system for ***glutamine*** ***synthetase*** of ***Mycobacterium*** tuberculosis together with a simple purification. The procedure allowed the structure of a complex with a phosphorylated form of the inhibitor methionine sulfoximine, magnesium, and ADP to be solved by molecular replacement and refined at 2.1-angstrom resolution. To our knowledge, this study provides the first reported structure for a taut form of the M. tuberculosis enzyme, similar to that observed for the Salmonella enzyme earlier. The phospho compound, generated in situ by an active enzyme, mimics the phosphorylated tetrahedral adduct at the transition state. Some differences in ligand interactions of the protein with both phosphorylated compound and nucleotide are observed compared with earlier structures; a third metal ion also is found. The importance of these differences in the catalytic mechanism is discussed; the results will help guide the search for specific inhibitors of potential therapeutic interest.
TI Structure of ***Mycobacterium*** tuberculosis ***glutamine*** ***synthetase*** in complex with a transition-state mimic provides functional insights.
AB ***Glutamine*** ***synthetase*** catalyzes the ligation of glutamate and ammonia to form glutamine, with the resulting hydrolysis of ATP. The enzyme is a central component of bacterial nitrogen metabolism and is a potential drug target. Here, we report a high-yield ***recombinant*** expression system for ***glutamine*** ***synthetase*** of ***Mycobacterium*** tuberculosis together with a simple purification. The procedure allowed the structure of a complex with a phosphorylated form of the. . .
IT . . .
Metabolism; Enzymology (Biochemistry and Molecular Biophysics)
IT Chemicals & Biochemicals
magnesium; glutamate; ammonia; nucleotides; ADP; glutamine; metal ion; ATP: hydrolysis; ***glutamine*** ***synthetase*** [EC 6.3.1.2]; methionine sulfoximine: enzyme inhibitor-drug; nitrogen: bacterial metabolism
ORGN . . .
Notes
Bacteria, Eubacteria, Microorganisms
ORGN Classifier
Mycobacteriaceae 08881
Super Taxa
Mycobacteria; Actinomycetes and Related Organisms; Eubacteria; Bacteria; Microorganisms
Organism Name
Mycobacterium tuberculosis (species): strain-H37Rv
Taxa Notes
Bacteria, Eubacteria, Microorganisms
RN 7439-95-4 (magnesium)
11070-68-1 (glutamate)
7664-41-7 (ammonia)
175832-20-9 (ADP)

6899-04-3 (glutamine)
111839-44-2 (ATP)
9023-70-5 (***glutamine*** ***synthetase***)
9023-70-5 (EC 6.3.1.2)
1982-67-8 (methionine sulfoximine)
7727-37-9 (nitrogen)

L10 ANSWER 10 OF 32 BIOSIS COPYRIGHT (c) 2009 The Thomson Corporation on STN DUPLICATE 4
AN 2005:498213 BIOSIS <<LOGINID::20090416>>
DN PREV200510285019
TI Analysis of the nearly identical morpholine monooxygenase-encoding mor genes from different ***Mycobacterium*** strains and characterization of the specific NADH : ferredoxin oxidoreductase of this cytochrome P450 system.
AU Sielaff, Bernhard; Andreesen, Jan R. [Reprint Author]
CS Univ Halle Wittenberg, Inst Mikrobiol, Kurt Mothes Str 3, D-06120 Halle, Germany
j.andreesen@mikrobiologie.uni-halle.de
SO Microbiology (Reading), (AUG 2005) Vol. 151, No. Part 8, pp. 2593-2603. ISSN: 1350-0872.
DT Article
LA English
ED Entered STN: 16 Nov 2005
Last Updated on STN: 16 Nov 2005
AB Cloning and sequencing of the morABC operon region revealed the genes encoding the three components of a cytochrome P450 monooxygenase, which is required for the degradation of the N-heterocycle morpholine by ***Mycobacterium*** sp. strain HE5. The cytochrome P450 (P450(mor)) and the Fe3S4 ferredoxin (Fd(mor)), encoded by morA and morB, respectively, have been characterized previously, whereas no evidence has hitherto been obtained for a specifically morpholine-induced reductase, which would be required to support the activity of the P450(mor) system. Analysis of the mor operon has now revealed the gene morC, encoding the ferredoxin reductase of this morpholine monooxygenase. The genes morA, morB and morC were identical to the corresponding genes from ***Mycobacterium*** sp. strain RP1. Almost identical mor genes in ***Mycobacterium*** chlorophenolicum PCP-1, in addition to an inducible cytochrome P450, pointing to horizontal gene transfer, were now identified. No evidence for a circular or linear plasmid was found in ***Mycobacterium*** sp. strain HE5. Analysis of the downstream sequences of morC revealed differences in this gene region between ***Mycobacterium*** sp. strain HE5 and ***Mycobacterium*** sp. strain RP1 on the one hand, and M. chlorophenolicum on the other hand, indicating insertions or deletions after recombination. Downstream of the mor genes, the gene orf1', encoding a putative ***glutamine*** ***synthetase*** , was identified in all studied strains. The gene morC of ***Mycobacterium*** sp. strain HE5 was heterologously expressed. The purified ***recombinant*** protein FdR(mor) was characterized as a monomeric 44 kDa protein, being a strictly NADH-dependent, FAD-containing reductase. The K-m values of FdR(mor) for the substrate NADH (37.7 +/- 4.1 mu M) and the artificial electron acceptors potassium ferricyanide (14.2 +/- 1.1 PM) and cytochrome c (28.0 +/- 3.6 mu M) were measured. FdR(mor) was shown to interact functionally with its natural redox partner, the Fe3S4 protein Fd(mor), and with the Fe2S2 protein adrenodoxin, albeit with a much lower efficiency, but not with spinach ferredoxin. In contrast, adrenodoxin

reductase, the natural redox partner of adrenodoxin, could not use Fd(mor) in activity assays. These results indicated that FdR(mor) can utilize different ferredoxins, but that Fd(mor) requires the specific NADH ferredoxin oxidoreductase FdR(mor) from the P450,or system for efficient catalytic function.

TI Analysis of the nearly identical morpholine monooxygenase-encoding mor genes from different ***Mycobacterium*** strains and characterization of the specific NADH : ferredoxin oxidoreductase of this cytochrome P450 system.

AB. . . encoding the three components of a cytochrome P450 monooxygenase, which is required for the degradation of the N-heterocycle morpholine by ***Mycobacterium*** sp. strain HE5. The cytochrome P450 (P450(mor)) and the Fe3S4 ferredoxin (Fd(mor)), encoded by morA and morB, respectively, have been. . . the ferredoxin reductase of this morpholine monooxygenase. The genes morA, morB and morC were identical to the corresponding genes from ***Mycobacterium*** sp. strain RP1. Almost identical mor genes in ***Mycobacterium*** chlorophenolicum PCP-1, in addition to an inducible cytochrome P450, pointing to horizontal gene transfer, were now identified. No evidence for a circular or linear plasmid was found in ***Mycobacterium*** sp. strain HE5. Analysis of the downstream sequences of morC revealed differences in this gene region between ***Mycobacterium*** sp. strain HE5 and ***Mycobacterium*** sp. strain RP1 on the one hand, and M. chlorophenolicum on the other hand, indicating insertions or deletions after recombination. Downstream of the mor genes, the gene orf1', encoding a putative ***glutamine*** ***synthetase*** , was identified in all studied strains. The gene morC of ***Mycobacterium*** sp. strain HE5 was heterologously expressed. The purified ***recombinant*** protein FdR(mor) was characterized as a monomeric 44 kDa protein, being a strictly NADH-dependent, FAD-containing reductase. The K-m values of. . .

IT . . . Concepts
Enzymology (Biochemistry and Molecular Biophysics); Molecular Genetics (Biochemistry and Molecular Biophysics)

IT Chemicals & Biochemicals
cytochrome c; potassium ferricyanide; ***glutamine*** ***synthetase*** [EC 6.3.1.2]; ferredoxin; cytochrome P450 monooxygenase [EC 1.14.14.1]; greigite; adrenodoxin; morABC operon; plasmid: linear, circular; NADH:ferredoxin oxidoreductase; morpholine monooxygenase

ORGN Classifier
Mycobacteriaceae 08881

Super Taxa
Mycobacteria; Actinomycetes and Related Organisms; Eubacteria; Bacteria; Microorganisms

Organism Name
Mycobacterium (genus): strain-HE5, strain-RP1
Mycobacterium chlorophenolicum (species): strain-PCP-1

Taxa Notes
Bacteria, Eubacteria, Microorganisms

RN 9007-43-6 (cytochrome c)
13746-66-2 (potassium ferricyanide)
9023-70-5 (***glutamine*** ***synthetase***)
9023-70-5 (EC 6.3.1.2)
9038-14-6 (cytochrome P450 monooxygenase)
9038-14-6 (EC 1.14.14.1)
12063-39-7 (greigite)

GEN ***Mycobacterium*** morC gene (Mycobacteriaceae): expression; ***Mycobacterium*** morA gene (Mycobacteriaceae); ***Mycobacterium*** morB gene (Mycobacteriaceae)

L10 ANSWER 11 OF 32 BIOSIS COPYRIGHT (c) 2009 The Thomson Corporation on STN DUPLICATE 5

AN 2005:522127 BIOSIS <<LOGINID::20090416>>

DN PREV200510307549

TI All four ***Mycobacterium*** tuberculosis glnA genes encode ***glutamine*** ***synthetase*** activities but only GlnA1 is abundantly expressed and essential for bacterial homeostasis.

AU Harth, Guenter; Maslesa-Galic, Sasa; Tullius, Michael V.; Horwitz, Marcus A. [Reprint Author]

CS Univ Calif Los Angeles, Sch Med, Dept Med, Div Infect Dis, 37-121 CHS,10833 Le Conte Ave, Los Angeles, CA 90095 USA
mhorwitz@mednet.ucla.edu

SO Molecular Microbiology, (NOV 2005) Vol. 58, No. 4, pp. 1157-1172.
CODEN: MOMIEE. ISSN: 0950-382X.

DT Article

LA English

ED Entered STN: 23 Nov 2005
Last Updated on STN: 23 Nov 2005

AB Glutamine synthetases (GS) are ubiquitous enzymes that play a central role in every cell's nitrogen metabolism. We have investigated the expression and activity of all four genomic ***Mycobacterium*** tuberculosis GS - GlnA1, GlnA2, GlnA3 and GlnA4 - and four enzymes regulating GS activity and/or nitrogen and glutamate metabolism - adenylyl transferase (GlnE), gamma-glutamylcysteine synthase (GshA), UDP-N-acetylmuramoylalanine-d-glutamate ligase (MurD) and glutamate racemase (MurI). All eight genes are located in multigene operons except for glnA1, and all are transcribed in M. tuberculosis; however, some are not translated or translated at such low levels that the enzymes escape detection. Of the four GS, only GlnA1 can be detected. Each of the eight genes, as well as the glnA1-glnE-glnA2 cluster, was expressed separately in ***Mycobacterium*** smegmatis, and its gene product was characterized and assayed for enzymatic activity by analysing the reaction products. In M. smegmatis, all four ***recombinant*** -overexpressed GS are multimeric enzymes exhibiting GS activity. Whereas GlnA1, GlnA3 and GlnA4 catalyse the synthesis of L-glutamine, GlnA2 catalyses the synthesis of D-glutamine and D-isoglutamine. The generation of mutants in M. tuberculosis of the four glnA genes, murD and murI demonstrated that all of these genes except glnA1 are nonessential for in vitro growth. L-methionine-S,R-sulphoximine (MSO), previously demonstrated to inhibit M. tuberculosis growth in vitro and in vivo, strongly inhibited all four GS enzymes; hence, the design of MSO analogues with an improved therapeutic to toxic ratio remains a promising strategy for the development of novel anti-M. tuberculosis drugs.

TI All four ***Mycobacterium*** tuberculosis glnA genes encode ***glutamine*** ***synthetase*** activities but only GlnA1 is abundantly expressed and essential for bacterial homeostasis.

AB. . . play a central role in every cell's nitrogen metabolism. We have investigated the expression and activity of all four genomic ***Mycobacterium*** tuberculosis GS - GlnA1, GlnA2, GlnA3 and GlnA4 - and four enzymes regulating GS activity and/or nitrogen and glutamate metabolism. . . only GlnA1 can be detected. Each of the eight genes, as well as the glnA1-glnE-glnA2 cluster, was expressed separately in ***Mycobacterium*** smegmatis, and its gene product was characterized

and assayed for enzymatic activity by analysing the reaction products. In M. smegmatis, all four ***recombinant*** -overexpressed GS are multimeric enzymes exhibiting GS activity. Whereas GlnA1, GlnA3 and GlnA4 catalyse the synthesis of L-glutamine, GlnA2 catalyses the. . .

IT Major Concepts
Pharmacology; Infection; Molecular Genetics (Biochemistry and Molecular Biophysics); Enzymology (Biochemistry and Molecular Biophysics)

IT Diseases
Mycobacterium tuberculosis infection: bacterial disease, infectious disease, drug therapy, genetics, pathology

IT Chemicals & Biochemicals
nitrogen: metabolism; gamma-glutamylcysteine synthase [EC 6.3.2.2];. . .

ORGN Classifier
Mycobacteriaceae 08881

Super Taxa
Mycobacteria; Actinomycetes and Related Organisms; Eubacteria; Bacteria; Microorganisms

Organism Name
Mycobacterium smegmatis (species)
Mycobacterium tuberculosis (species)

Taxa Notes
Bacteria, Eubacteria, Microorganisms

GEN ***Mycobacterium*** tuberculosis GlnA1 gene (Mycobacteriaceae): expression, bacterial homeostasis; ***Mycobacterium*** tuberculosis GlnA2 gene (Mycobacteriaceae): expression; ***Mycobacterium*** tuberculosis GlnA3 gene (Mycobacteriaceae): expression; ***Mycobacterium*** tuberculosis GlnA4 gene (Mycobacteriaceae): expression; ***Mycobacterium*** tuberculosis GlnE gene (Mycobacteriaceae); ***Mycobacterium*** tuberculosis GshA gene (Mycobacteriaceae); ***Mycobacterium*** tuberculosis MurD gene (Mycobacteriaceae); ***Mycobacterium*** tuberculosis Murl gene (Mycobacteriaceae)

L10 ANSWER 12 OF 32 BIOSIS COPYRIGHT (c) 2009 The Thomson Corporation on STN DUPLICATE 6

AN 2005:503424 BIOSIS <<LOGINID::20090416>>

DN PREV200510280522

TI Development of a simple assay protocol for high-throughput screening of ***Mycobacterium*** tuberculosis ***glutamine*** ***synthetase*** for the identification of novel inhibitors.

AU Singh, Upasana; Panchanadikar, Vinita; Sarkar, Dhiman [Reprint Author]

CS Natl Chem Lab, Combichem Bioresource Ctr, Dr Homi Bhabha Rd, Poona 411008, Maharashtra, India
dsarkar@dalton.ncl.res.in

SO Journal of Biomolecular Screening, (OCT 2005) Vol. 10, No. 7, pp. 725-729. ISSN: 1087-0571.

DT Article

LA English

ED Entered STN: 16 Nov 2005
Last Updated on STN: 16 Nov 2005

AB ***Mycobacterium*** tuberculosis ***glutamine*** ***synthetase*** (GS) is an essential enzyme involved in the pathogenicity of the organism. The screening of a compound library using a robust high-throughput screening (HTS) assay is currently thought to be the most efficient way of getting lead molecules, which are potent inhibitors for this enzyme. The authors have purified the enzyme to a >

90% level from the ***recombinant*** Escherichia coli strain YMC21E, and it was used for partial characterization as well as standardization experiments. The results indicated that the K-m of the enzyme for L-glutamine and hydroxylamine were 60 mM and 8.3 mM, respectively. The K-m for ADP, arsenate, and Mn2+, were 2 mu M, 5 mu M, and 25 mu M, respectively. When the components were adjusted according to their K-m values, the activity remained constant for at least 3 h at both 25 degrees C and 37 degrees C. The Z' factor determined in microplate format indicated robustness of the assay. When the signal/noise ratios were determined for different assay volumes, it was observed that the 200-mu l volume was found to be optimum. The DMSO tolerance of the enzyme was checked up to 10%, with minimal inhibition. The IC50 value determined for L-methionine S-sulfoximine on the enzyme activity was 3 mM. Approximately 18,000 small molecules could be screened per day using this protocol by a Beckman Coulter HTS setup.

TI Development of a simple assay protocol for high-throughput screening of ***Mycobacterium*** tuberculosis ***glutamine*** ***synthetase*** for the identification of novel inhibitors.

AB ***Mycobacterium*** tuberculosis ***glutamine*** ***synthetase*** (GS) is an essential enzyme involved in the pathogenicity of the organism. The screening of a compound library using a. . . which are potent inhibitors for this enzyme. The authors have purified the enzyme to a > 90% level from the ***recombinant*** Escherichia coli strain YMC21E, and it was used for partial characterization as well as standardization experiments. The results indicated that. . .

IT Major Concepts
Methods and Techniques; Enzymology (Biochemistry and Molecular Biophysics)

IT Chemicals & Biochemicals
hydroxylamine; ***glutamine*** ***synthetase*** [EC 6.3.1.2]; L-glutamine

ORGN . . .

Notes
Bacteria, Eubacteria, Microorganisms

ORGN Classifier
Mycobacteriaceae 08881

Super Taxa
Mycobacteria; Actinomycetes and Related Organisms; Eubacteria; Bacteria; Microorganisms

Organism Name
Mycobacterium tuberculosis (species): pathogen

Taxa Notes
Bacteria, Eubacteria, Microorganisms

RN 7803-49-8 (hydroxylamine)
9023-70-5 (***glutamine*** ***synthetase***)
9023-70-5 (EC 6.3.1.2)
56-85-9 (L-glutamine)

L10 ANSWER 13 OF 32 SCISEARCH COPYRIGHT (c) 2009 The Thomson Corporation on STN

AN 2004:684567 SCISEARCH <<LOGINID::20090416>>

GA The Genuine Article (R) Number: 840KQ

TI A two-plasmid system for stable, selective-pressure-independent expression of multiple extracellular proteins in mycobacteria

AU Horwitz M A (Reprint)

CS Univ Calif Los Angeles, Dept Med, Div Infect Dis, 37-121 CHS, 10833 Le

Conte Ave, Los Angeles, CA 90095 USA (Reprint)
AU Harth G; Maslesa-Galic S
CS Univ Calif Los Angeles, Dept Med, Div Infect Dis, Los Angeles, CA 90095 USA
E-mail: mhorwitz@mednet.ucla.edu
CYA USA
SO MICROBIOLOGY-SGM, (JUL 2004) Vol. 150, Part 7, pp. 2143-2151.
ISSN: 1350-0872.
PB SOC GENERAL MICROBIOLOGY, MARLBOROUGH HOUSE, BASINGSTOKE RD, SPENCERS WOODS, READING RG7 1AG, BERKS, ENGLAND.
DT Article; Journal
LA English
REC Reference Count: 27
ED Entered STN: 20 Aug 2004
Last Updated on STN: 20 Aug 2004
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS
AB ***Recombinant*** mycobacteria expressing ***Mycobacterium*** tuberculosis extracellular proteins are leading candidates for new vaccines against tuberculosis and other mycobacterial diseases, and important tools both in anti mycobacterial drug development and basic research in mycobacterial pathogenesis. ***Recombinant*** mycobacteria that stably overexpress and secrete major extracellular proteins of M. tuberculosis in native form on plasmids pSMT3 and pNBV1 were previously constructed by the authors. To enhance the versatility of this plasmid-based approach for mycobacterial protein expression, the Escherichia coli/mycobacteria shuttle plasmid pGB9 was modified to accommodate mycobacterial genes expressed from their endogenous promoters. Previous studies showed that the modified plasmid, designated pGB9.2, derived from the cryptic ***Mycobacterium*** fortuitum plasmid pMF1, was present at a low copy number in both E. coli and mycobacteria, and expression of ***recombinant*** M. tuberculosis proteins was found to be at levels paralleling its copy number, that is, approximating their endogenous levels. Plasmid pGB9.2 was compatible with the shuttle vectors pSMT3 and pNBV1 and in combination with them it simultaneously expressed the M. tuberculosis 30 kDa extracellular protein FbpB. Plasmid pGB9.2 was stably maintained in the absence of selective pressure in three mycobacterial species: ***Mycobacterium*** bovis BCG, M. tuberculosis and M. smegmatis. Plasmid pGB9.2 was found to be self-transmissible between both fast- and slow-growing mycobacteria, but not from mycobacteria to E. coli or between E. coli strains. The combination of two compatible plasmids in one BCG strain allows expression of ***recombinant*** mycobacterial proteins at different levels, a potentially important factor in optimizing vaccine potency.
AB ***Recombinant*** mycobacteria expressing ***Mycobacterium*** tuberculosis extracellular proteins are leading candidates for new vaccines against tuberculosis and other mycobacterial diseases, and important tools both in anti mycobacterial drug development and basic research in mycobacterial pathogenesis. ***Recombinant*** mycobacteria that stably overexpress and secrete major extracellular proteins of M. tuberculosis in native form on plasmids pSMT3 and pNBV1. . . mycobacterial genes expressed from their endogenous promoters. Previous studies showed that the modified plasmid, designated pGB9.2, derived from the cryptic ***Mycobacterium*** fortuitum plasmid pMF1, was present at a low copy number in both E. coli and mycobacteria, and expression of ***recombinant*** M. tuberculosis proteins was found to be at levels paralleling its copy number, that is, approximating their endogenous levels. Plasmid. . . 30 kDa extracellular protein FbpB.

Plasmid pGB9.2 was stably maintained in the absence of selective pressure in three mycobacterial species: ***Mycobacterium*** bovis BCG, M. tuberculosis and M. smegmatis. Plasmid pGB9.2 was found to be self-transmissible between both fast- and slow-growing mycobacteria,. . . E. coli or between E. coli strains. The combination of two compatible plasmids in one BCG strain allows expression of ***recombinant*** mycobacterial proteins at different levels, a potentially important factor in optimizing vaccine potency.

STP KeyWords Plus (R): HYGROMYCIN-RESISTANCE; ***GLUTAMINE*** - ***SYNTHETASE*** ; PROTECTIVE IMMUNITY; MYCOLYL TRANSFERASE; TUBERCULOSIS; PLASMID; INHIBITORS; SECRETION; FORTUITUM; ANTIGEN

L10 ANSWER 14 OF 32 SCISEARCH COPYRIGHT (c) 2009 The Thomson Corporation on STN

AN 2004:538882 SCISEARCH <<LOGINID::20090416>>

GA The Genuine Article (R) Number: 826YV

TI The ***Mycobacterium*** tuberculosis protein serine/threonine kinase PknG is linked to cellular glutamate/glutamine levels and is important for growth in vivo

AU Av-Gay Y (Reprint)

CS Univ British Columbia, Dept Med, Div Infect Dis, 2733 Heather St, Vancouver, BC V5Z 3J5, Canada (Reprint)

AU Cowley S; Ko M; Pick N; Chow R; Downing K J; Gordhan B G; Betts J C; Mizrahi V; Smith D A; Stokes R W

CS Univ British Columbia, Dept Med, Div Infect Dis, Vancouver, BC V5Z 3J5, Canada; NHLS, Mol Mycobacteriol Res Unit, Johannesburg, South Africa; Univ Witwatersrand, Johannesburg, South Africa; GlaxoSmithKline, Stevenage, Herts, England; London Sch Hyg & Trop Med, London WC1, England; Univ British Columbia, Dept Pediat, Vancouver, BC V6T 1W5, Canada

CYA Canada; South Africa; England

SO MOLECULAR MICROBIOLOGY, (JUN 2004) Vol. 52, No. 6, pp. 1691-1702. ISSN: 0950-382X.

PB BLACKWELL PUBLISHING LTD, 9600 GARSINGTON RD, OXFORD OX4 2DG, OXON, ENGLAND.

DT Article; Journal

LA English

REC Reference Count: 48

ED Entered STN: 2 Jul 2004
Last Updated on STN: 2 Jul 2004

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB The function of the ***Mycobacterium*** tuberculosis eukaryotic-like protein serine/threonine kinase PknG was investigated by gene knock-out and by expression and biochemical analysis. The pknG gene (Rv0410c), when cloned and expressed in Escherichia coli, encodes a functional kinase. An in vitro kinase assay of the ***recombinant*** protein demonstrated that PknG can autophosphorylate its kinase domain as well as its 30 kDa C-terminal portion, which contains a tetratricopeptide (TPR) structural signalling motif. Western analysis revealed that PknG is located in the cytosol as well as in mycobacterial membrane. The pknG gene was inactivated by allelic exchange in M. tuberculosis. The resulting mutant strain causes delayed mortality in SCID mice and displays decreased viability both in vitro and upon infection of BALB/c mice. The reduced growth of the mutant was more pronounced in the stationary phase of the mycobacterial growth cycle and when grown in nutrient-depleted media. The PknG-deficient mutant accumulates glutamate and glutamine. The cellular levels of these two amino acids reached approximately threefold of their parental strain levels. Higher cellular levels of the

amine sugar-containing molecules, GlcN-Ins and mycothiol, which are derived from glutamate, were detected in the DeltapknG mutant. De novo glutamine synthesis was shown to be reduced by 50%. This is consistent with current knowledge suggesting that glutamine synthesis is regulated by glutamate and glutamine levels. These data support our hypothesis that PknG mediates the transfer of signals sensing nutritional stress in M. tuberculosis and translates them into metabolic adaptation.

TI The ***Mycobacterium*** tuberculosis protein serine/threonine kinase PknG is linked to cellular glutamate/glutamine levels and is important for growth in vivo

AB The function of the ***Mycobacterium*** tuberculosis eukaryotic-like protein serine/threonine kinase PknG was investigated by gene knock-out and by expression and biochemical analysis. The pknG gene (Rv0410c), when cloned and expressed in Escherichia coli, encodes a functional kinase. An in vitro kinase assay of the ***recombinant*** protein demonstrated that PknG can autophosphorylate its kinase domain as well as its 30 kDa C-terminal portion, which contains a. . .

STP KeyWords Plus (R): ***GLUTAMINE*** - ***SYNTHETASE*** ; HOMOLOGOUS RECOMBINATION; BACILLUS-SUBTILIS; GENE REPLACEMENT; GUINEA-PIGS; EXPRESSION; MICE; RESISTANCE; MYCOTHIOL; INFECTION

L10 ANSWER 15 OF 32 BIOSIS COPYRIGHT (c) 2009 The Thomson Corporation on STN DUPLICATE 7

AN 2004:271517 BIOSIS <<LOGINID::20090416>>

DN PREV200400271160

TI Cloning and expression of mycobacterial ***glutamine*** ***synthetase*** gene in Escherichia coli.

AU Singh, Jitendra; Joshi, Mohan Chandra; Bhatnagar, Rakesh [Reprint Author]

CS Ctr Biotechnol, Jawaharlal Nehru Univ, New Delhi, 110067, India
rakeshbhatnagar@mail.jnu.ac.in

SO Biochemical and Biophysical Research Communications, (April 30 2004) Vol. 317, No. 2, pp. 634-638. print.
CODEN: BBRCA9. ISSN: 0006-291X.

DT Article

LA English

ED Entered STN: 26 May 2004
Last Updated on STN: 26 May 2004

AB Extracellular ***glutamine*** ***synthetase*** (GS) is one of the prominent proteins secreted by pathogenic mycobacteria such as ***Mycobacterium*** tuberculosis and ***Mycobacterium*** bovis. Non-pathogenic species like ***Mycobacterium*** smegmatis and ***Mycobacterium*** phlei do not secrete this protein. GS has been proposed to play an indispensable role in intracellular survival of pathogenic mycobacteria. In this study, the structural gene for extracellular ***glutamine*** ***synthetase*** of M. tuberculosis was PCR amplified and expressed as fusion protein with hexahistidine residues in Escherichia coli. Expression of GS in E. coli under transcriptional regulation of T5 promoter yielded an insoluble protein aggregating to form inclusion bodies. The inclusion bodies were solubilized in presence of 8 M urea and the enzyme was purified to homogeneity under denaturing conditions using nitrilotriacetic acid (Ni-NTA) affinity chromatography. The denatured protein was renatured by gradual removal of the urea while immobilized on (Ni-NTA) column. The yield of purified ***recombinant*** ***glutamine*** ***synthetase*** was 40mg/L. The purified ***recombinant*** enzyme was obtained in highly active state having specific activity of 200 U/mg protein. This is the first report describing cloning and expression of

mycobacterial ***glutamine*** ***synthetase*** gene in E. coli.
Copyright 2004 Elsevier Inc. All rights reserved.
TI Cloning and expression of mycobacterial ***glutamine*** ***synthetase*** gene in Escherichia coli.
AB Extracellular ***glutamine*** ***synthetase*** (GS) is one of the prominent proteins secreted by pathogenic mycobacteria such as ***Mycobacterium*** tuberculosis and ***Mycobacterium*** bovis. Non-pathogenic species like ***Mycobacterium*** smegmatis and ***Mycobacterium*** phlei do not secrete this protein. GS has been proposed to play an indispensable role in intracellular survival of pathogenic mycobacteria. In this study, the structural gene for extracellular ***glutamine*** ***synthetase*** of M. tuberculosis was PCR amplified and expressed as fusion protein with hexahistidine residues in Escherichia coli. Expression of GS. . . The denatured protein was renatured by gradual removal of the urea while immobilized on (Ni-NTA) column. The yield of purified ***recombinant*** ***glutamine*** ***synthetase*** was 40mg/L. The purified ***recombinant*** enzyme was obtained in highly active state having specific activity of 200 U/mg protein. This is the first report describing cloning and expression of mycobacterial ***glutamine*** ***synthetase*** gene in E. coli. Copyright 2004 Elsevier Inc. All rights reserved.
IT Major Concepts
Molecular Genetics (Biochemistry and Molecular Biophysics)
IT Chemicals & Biochemicals
glutamine ***synthetase*** [EC 6.3.1.2]; mycobacterial ***glutamine*** ***synthetase***
ORGN . . .
Notes
Bacteria, Eubacteria, Microorganisms
ORGN Classifier
Mycobacteriaceae 08881
Super Taxa
Mycobacteria; Actinomycetes and Related Organisms; Eubacteria; Bacteria; Microorganisms
Organism Name
Mycobacterium bovis (species): pathogen
Mycobacterium tuberculosis (species): pathogen
Taxa Notes
Bacteria, Eubacteria, Microorganisms
RN 9023-70-5 (***glutamine*** ***synthetase***)
9023-70-5 (EC 6.3.1.2)
GEN Escherichia coli GS gene [Escherichia coli ***glutamine*** ***synthetase*** gene] (Enterobacteriaceae); human mycobacterial ***glutamine*** ***synthetase*** gene (Hominidae): cloning, expression

L10 ANSWER 16 OF 32 CAPLUS COPYRIGHT 2009 ACS on STN
AN 2004:94959 CAPLUS <<LOGINID::20090416>>
DN 141:2030
TI Molecular cloning, nucleotide sequencing and expression of genes encoding a cytochrome P450 system involved in secondary amine utilization in ***Mycobacterium*** sp. strain RP1
AU Trigui, Mohamed; Pulvin, Sylviane; Truffaut, Nicole; Thomas, Daniel; Poupin, Pascal
CS MR 6022 CNRS, Laboratoire de Technologie Enzymatique, Universite de Technologie de Compiegne, Compiegne, 60205, Fr.

SO Research in Microbiology (2004), 155(1), 1-9
CODEN: RMCREW; ISSN: 0923-2508
PB Elsevier Science B.V.
DT Journal
LA English
AB ***Mycobacterium*** sp. strain RP1 degrades morpholine, piperidine, and pyrrolidine and is able to use these compds. as the sole source of carbon, nitrogen, and energy. Cytochrome P 450 (MorA) is involved in the biodegrdn. of these secondary amines. A 3.9-PstI genomic DNA fragment, contg. the gene encoding MorA, was cloned and sequenced. Four open reading frames were detected on this DNA fragment. The first encoded a cytochrome P 450 designated as MorA which was the second member of the CYP151 family and was named CYP151A2. The second open reading frame (morB) featured a [3Fe-4S] type of ferredoxin. A third gene (morC), exhibiting sequence identity to known reductases, and a fourth truncated gene encoding a putative glutamine reductase (orf 1'), were found downstream of morB. ***Recombinant*** MorA cytochrome P 450 was purified to homogeneity from Escherichia coli. The purified enzyme was a monomeric sol. protein with an apparent Mr of about 45,000. CYP151A2 catalyzed the ring cleavage of the secondary amines and the Vmax/KMapp values indicated that pyrrolidine is the preferred substrate for this monooxygenase.
RE.CNT 47 THERE ARE 47 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT
TI Molecular cloning, nucleotide sequencing and expression of genes encoding a cytochrome P450 system involved in secondary amine utilization in ***Mycobacterium*** sp. strain RP1
AB ***Mycobacterium*** sp. strain RP1 degrades morpholine, piperidine, and pyrrolidine and is able to use these compds. as the sole source of. . . to known reductases, and a fourth truncated gene encoding a putative glutamine reductase (orf 1'), were found downstream of morB. ***Recombinant*** MorA cytochrome P 450 was purified to homogeneity from Escherichia coli. The purified enzyme was a monomeric sol. protein with. . .
ST cytochrome CYP151A2 gene MorA sequence ***Mycobacterium*** secondary amine degrdn; sequence ***Mycobacterium*** MorB MorC ***glutamine*** ***synthetase*** gene
IT Ferredoxins
RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
(gene morB, sequence homolog; mol. cloning, nucleotide sequencing and expression of genes encoding a cytochrome P 450 system involved in secondary amine utilization in ***Mycobacterium*** sp. strain RP1)
IT DNA sequences
Mycobacterium
Protein sequences
(mol. cloning, nucleotide sequencing and expression of genes encoding a cytochrome P 450 system involved in secondary amine utilization in ***Mycobacterium*** sp. strain RP1)
IT Gene, microbial
RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
(morA; mol. cloning, nucleotide sequencing and expression of genes encoding a cytochrome P 450 system involved in secondary amine utilization in ***Mycobacterium*** sp. strain RP1)
IT Gene, microbial

RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
(morB; mol. cloning, nucleotide sequencing and expression of genes encoding a cytochrome P 450 system involved in secondary amine utilization in ***Mycobacterium*** sp. strain RP1)

IT Gene, microbial
RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
(morC; mol. cloning, nucleotide sequencing and expression of genes encoding a cytochrome P 450 system involved in secondary amine utilization in ***Mycobacterium*** sp. strain RP1)

IT 487549-48-4P
RL: BPN (Biosynthetic preparation); PRP (Properties); PUR (Purification or recovery); BIOL (Biological study); PREP (Preparation)
(amino acid sequence; mol. cloning, nucleotide sequencing and expression of genes encoding a cytochrome P 450 system involved in secondary amine utilization in ***Mycobacterium*** sp. strain RP1)

IT 487549-50-8 487549-51-9 487549-52-0
RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
(amino acid sequence; mol. cloning, nucleotide sequencing and expression of genes encoding a cytochrome P 450 system involved in secondary amine utilization in ***Mycobacterium*** sp. strain RP1)

IT 110-89-4, Piperidine, reactions 110-91-8, Morpholine, reactions
123-75-1, Pyrrolidine, reactions
RL: RCT (Reactant); RACT (Reactant or reagent)
(gene morA monooxygenase substrate; mol. cloning, nucleotide sequencing and expression of genes encoding a cytochrome P 450 system involved in secondary amine utilization in ***Mycobacterium*** sp. strain RP1)

IT 9035-51-2, Cytochrome P 450, biological studies 9038-14-6, Monooxygenase
RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
(gene morA; mol. cloning, nucleotide sequencing and expression of genes encoding a cytochrome P 450 system involved in secondary amine utilization in ***Mycobacterium*** sp. strain RP1)

IT 9029-33-8, Ferredoxin-NADP reductase
RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
(gene morC, sequence homolog; mol. cloning, nucleotide sequencing and expression of genes encoding a cytochrome P 450 system involved in secondary amine utilization in ***Mycobacterium*** sp. strain RP1)

IT 417982-38-8
RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
(nucleotide sequence; mol. cloning, nucleotide sequencing and expression of genes encoding a cytochrome P 450 system involved in secondary amine utilization in ***Mycobacterium*** sp. strain RP1)

IT 9023-70-5, ***Glutamine*** ***synthetase***
RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
(sequence homolog; mol. cloning, nucleotide sequencing and expression of genes encoding a cytochrome P 450 system involved in secondary amine utilization in ***Mycobacterium*** sp. strain RP1)

L10 ANSWER 17 OF 32 CAPLUS COPYRIGHT 2009 ACS on STN
AN 2003:855955 CAPLUS <<LOGINID::20090416>>
DN 139:363579

TI Tuberculosis vaccines including ***recombinant*** ***Mycobacterium*** bovis-BCG strains expressing ***alanine*** ***dehydrogenase*** , ***serine*** ***dehydratase*** and/or ***glutamine*** ***synthetase***

IN Liu, Jun; Chen, Jeffrey; Alexander, David

PA Can.

SO PCT Int. Appl., 78 pp.
CODEN: PIXXD2

DT Patent

LA English

FAN.CNT 1

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	WO 2003089462	A2	20031030	WO 2003-CA566	20030416
	WO 2003089462	A3	20040521		
	W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW				
	RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
	CA 2481108	A1	20031030	CA 2003-2481108	20030416
	AU 2003218838	A1	20031103	AU 2003-218838	20030416
	GB 2403477	A	20050105	GB 2004-25165	20030416
	GB 2403477	B	20060823		
	CN 1703513	A	20051130	CN 2003-802276	20030416
	JP 2006508633	T	20060316	JP 2003-586182	20030416
	JP 4233458	B2	20090304		
	RU 2339692	C2	20081127	RU 2004-133751	20030416
	ZA 2004008344	A	20050907	ZA 2004-8344	20041014
	US 20070264286	A1	20071115	US 2006-511718	20060728
PRAI	US 2002-372450P	P	20020416		
	WO 2003-CA566	W	20030416		

AB The invention relates to a live ***recombinant*** ***Mycobacterium*** bovis-BCG strain comprising a nucleic acid capable of expression, the nucleic acid encoding at least one protein or polypeptide that exhibits ***alanine*** ***dehydrogenase*** activity, ***glutamine*** ***synthetase*** activity, or ***serine*** ***dehydratase*** activity. The ***recombinant*** ***alanine*** ***dehydrogenase*** , ***serine*** ***dehydratase*** and ***glutamine*** ***synthetase*** are derived from ***Mycobacterium*** tuberculosis.

RE.CNT 3 THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

TI Tuberculosis vaccines including ***recombinant*** ***Mycobacterium*** bovis-BCG strains expressing ***alanine*** ***dehydrogenase*** , ***serine*** ***dehydratase*** and/or ***glutamine*** ***synthetase***

AB The invention relates to a live ***recombinant*** ***Mycobacterium*** bovis-BCG strain comprising a nucleic acid capable of expression, the nucleic acid encoding at least one protein or polypeptide that exhibits ***alanine*** ***dehydrogenase*** activity, ***glutamine*** ***synthetase*** activity, or

serine ***dehydratase*** activity. The ***recombinant*** ***alanine*** ***dehydrogenase*** , ***serine*** ***dehydratase*** and ***glutamine*** ***synthetase*** are derived from ***Mycobacterium*** tuberculosis.

ST ***recombinant*** ***Mycobacterium*** bovis BCG strain tuberculosis vaccine; ***alanine*** ***dehydrogenase*** ***serine*** ***dehydratase*** ***glutamine*** ***synthetase*** BCG tuberculosis vaccine

IT Immunostimulants
(adjuvants; tuberculosis vaccines including ***recombinant*** ***Mycobacterium*** bovis-BCG strains expressing ***alanine*** ***dehydrogenase*** , ***serine*** ***dehydratase*** and/or ***glutamine*** ***synthetase***)

IT Drug delivery systems
(carriers; tuberculosis vaccines including ***recombinant*** ***Mycobacterium*** bovis-BCG strains expressing ***alanine*** ***dehydrogenase*** , ***serine*** ***dehydratase*** and/or ***glutamine*** ***synthetase***)

IT Proteins
RL: BPN (Biosynthetic preparation); BSU (Biological study, unclassified); PRP (Properties); THU (Therapeutic use); BIOL (Biological study); PREP (Preparation); USES (Uses)
(***recombinant*** ; tuberculosis vaccines including ***recombinant*** ***Mycobacterium*** bovis-BCG strains expressing ***alanine*** ***dehydrogenase*** , ***serine*** ***dehydratase*** and/or ***glutamine*** ***synthetase***)

IT Antitumor agents
Bladder, neoplasm
Bos taurus
Culture media
DNA sequences
Human
Mammalia
Molecular cloning
Mycobacterium
Mycobacterium BCG
Mycobacterium tuberculosis
Pathogen
Protein sequences
Test kits
Tuberculosis
Vaccines
(tuberculosis vaccines including ***recombinant*** ***Mycobacterium*** bovis-BCG strains expressing ***alanine*** ***dehydrogenase*** , ***serine*** ***dehydratase*** and/or ***glutamine*** ***synthetase***)

IT Gene, microbial
Nucleic acids
RL: BPN (Biosynthetic preparation); BSU (Biological study, unclassified); PRP (Properties); THU (Therapeutic use); BIOL (Biological study); PREP (Preparation); USES (Uses)
(tuberculosis vaccines including ***recombinant*** ***Mycobacterium*** bovis-BCG strains expressing ***alanine*** ***dehydrogenase*** , ***serine*** ***dehydratase*** and/or ***glutamine*** ***synthetase***)

IT Antigens
RL: BSU (Biological study, unclassified); THU (Therapeutic use); BIOL

(Biological study); USES (Uses)
(tuberculosis vaccines including ***recombinant*** ***Mycobacterium*** bovis-BCG strains expressing ***alanine*** ***dehydrogenase*** , ***serine*** ***dehydratase*** and/or ***glutamine*** ***synthetase***)

IT 619345-18-5P 619345-20-9P 619345-21-0P 619345-22-1P 619345-23-2P 619345-24-3P
RL: BPN (Biosynthetic preparation); BSU (Biological study, unclassified); PRP (Properties); THU (Therapeutic use); BIOL (Biological study); PREP (Preparation); USES (Uses)
(amino acid sequence; tuberculosis vaccines including ***recombinant*** ***Mycobacterium*** bovis-BCG strains expressing ***alanine*** ***dehydrogenase*** , ***serine*** ***dehydratase*** and/or ***glutamine*** ***synthetase***)

IT 619345-19-6
RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
(amino acid sequence; tuberculosis vaccines including ***recombinant*** ***Mycobacterium*** bovis-BCG strains expressing ***alanine*** ***dehydrogenase*** , ***serine*** ***dehydratase*** and/or ***glutamine*** ***synthetase***)

IT 619345-25-4P 619345-27-6P 619345-28-7P 619345-29-8P 619345-30-1P 619345-31-2P
RL: BPN (Biosynthetic preparation); BSU (Biological study, unclassified); PRP (Properties); THU (Therapeutic use); BIOL (Biological study); PREP (Preparation); USES (Uses)
(nucleotide sequence; tuberculosis vaccines including ***recombinant*** ***Mycobacterium*** bovis-BCG strains expressing ***alanine*** ***dehydrogenase*** , ***serine*** ***dehydratase*** and/or ***glutamine*** ***synthetase***)

IT 619345-26-5, DNA (***Mycobacterium*** bovis gene ald)
RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
(nucleotide sequence; tuberculosis vaccines including ***recombinant*** ***Mycobacterium*** bovis-BCG strains expressing ***alanine*** ***dehydrogenase*** , ***serine*** ***dehydratase*** and/or ***glutamine*** ***synthetase***)

IT 7440-44-0, Carbon, biological studies 7727-37-9, Nitrogen, biological studies
RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(source; tuberculosis vaccines including ***recombinant*** ***Mycobacterium*** bovis-BCG strains expressing ***alanine*** ***dehydrogenase*** , ***serine*** ***dehydratase*** and/or ***glutamine*** ***synthetase***)

IT 9014-27-1P, ***Serine*** ***dehydratase*** 9023-70-5P, ***Glutamine*** ***synthetase*** 9029-06-5P, ***Alanine*** ***dehydrogenase*** 175380-16-2P, GenBank Z70692 193398-67-3P, GenBank Z97193 196526-70-2P, GenBank U87280 199902-12-0P, GenBank AL008883 202943-88-2P, GenBank AL021428 335511-06-3P, GenBank AE006919 335512-36-2P, GenBank AE007049 335512-60-2P, GenBank AE007073 335513-04-7P, GenBank AE007117
RL: BPN (Biosynthetic preparation); BSU (Biological study, unclassified); PRP (Properties); THU (Therapeutic use); BIOL (Biological study); PREP (Preparation); USES (Uses)
(tuberculosis vaccines including ***recombinant*** ***Mycobacterium*** bovis-BCG strains expressing ***alanine***

dehydrogenase , ***serine*** ***dehydratase*** and/or ***glutamine*** ***synthetase***)

IT 50-99-7, Dextrose, biological studies 56-41-7, L-Alanine, biological studies 56-45-1, L-Serine, biological studies 56-81-5, Glycerol, biological studies 71-00-1, L-Histidine, biological studies 77-92-9, Citric acid, biological studies 338-69-2, D-Alanine 7439-89-6, Iron, biological studies 7439-95-4, Magnesium, biological studies 14808-79-8, Sulfate, biological studies
RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(tuberculosis vaccines including ***recombinant*** ***Mycobacterium*** bovis-BCG strains expressing ***alanine*** ***dehydrogenase*** , ***serine*** ***dehydratase*** and/or ***glutamine*** ***synthetase***)

L10 ANSWER 18 OF 32 SCISEARCH COPYRIGHT (c) 2009 The Thomson Corporation on STN
AN 2003:1051894 SCISEARCH <<LOGINID::20090416>>
GA The Genuine Article (R) Number: 747ZA
TI The ***Mycobacterium*** tuberculosis complex-restricted gene cfp32 encodes an expressed protein that is detectable in tuberculosis patients and is positively correlated with pulmonary interleukin-10
AU Ho J L (Reprint)
CS Cornell Univ, Joan & Sanford I Weill Med Coll, Dept Med, Div Int Med & Infect Dis, Room A-421, 525 E 68th St, New York, NY 10021 USA (Reprint)
AU Huard R C; Chitale S; Leung M; Lazzarini L C O; Zhu H X; Shashkina E; Laal S; Conde M B; Kritski A L; Belisle J T; Kreiswirth B N; Silva J R L E
CS Cornell Univ, Joan & Sanford I Weill Med Coll, Dept Med, Div Int Med & Infect Dis, New York, NY 10021 USA; Cornell Univ, Grad Sch Med, New York, NY USA; NYU, Sch Med, Dept Pathol, New York, NY USA; Vet Affairs Med Ctr, Res Ctr AIDS & HIV Invect, New York, NY USA; Univ Med & Dent New Jersey, New Jersey Med Sch, Natl TB Ctr, Newark, NJ 07103 USA; Univ Fed Rio de Janeiro, Hosp Univ Clementino Fraga Filho, Inst Doencas Torax, Rio De Janeiro, Brazil; Colorado State Univ, Dept Microbiol Immunol & Pathol, Mycobacteria Res Labs, Ft Collins, CO 80523 USA
CYA USA; Brazil
SO INFECTION AND IMMUNITY, (DEC 2003) Vol. 71, No. 12, pp. 6871-6883. ISSN: 0019-9567.
PB AMER SOC MICROBIOLOGY, 1752 N ST NW, WASHINGTON, DC 20036-2904 USA.
DT Article; Journal
LA English
REC Reference Count: 74
ED Entered STN: 12 Dec 2003
Last Updated on STN: 12 Dec 2003
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS
AB Human tuberculosis (TB) is caused by the bacillus ***Mycobacterium*** tuberculosis, a subspecies of the M. tuberculosis complex (MTC) of mycobacteria. Postgenomic dissection of the M. tuberculosis proteome is ongoing and critical to furthering our understanding of factors mediating M. tuberculosis pathobiology. Towards this end, a 32-kDa putative glyoxalase in the culture filtrate (CF) of growing M. tuberculosis (originally annotated as Rv0577 and hereafter designated CFP32) was identified, cloned, and characterized. The cfp32 gene is MTC restricted, and the gene product is expressed ex vivo as determined by the respective Southern and Western blot testing of an assortment of mycobacteria. Moreover, the cfp32 gene sequence is conserved within the MTC, as no polymorphisms were found in the tested cfp32 PCR products upon sequence

analysis. Western blotting of M. tuberculosis subcellular fractions localized CFP32 predominantly to the CF and cytosolic compartments. Data to support the in vivo expression of CFP32 were provided by the serum recognition of ***recombinant*** CFP32 in 32% of TB patients by enzyme-linked immunosorbent assay (ELISA) as well as the direct detection of CFP32 by ELISA in the induced sputum samples from 56% of pulmonary TB patients. Of greatest interest was the observation that, per sample, sputum CFP32 levels (a potential indicator of increasing bacterial burden) correlated with levels of expression in sputum of interleukin-10 (an immunosuppressive cytokine and a putative contributing factor to disease progression) but not levels of gamma interferon (a key cytokine in the protective immune response in TB), as measured by ELISA. Combined, these data suggest that CFP32 serves a necessary biological function(s) in tubercle bacilli and may contribute to the M. tuberculosis pathogenic mechanism. Overall, CFP32 is an attractive target for drug and vaccine design as well as new diagnostic strategies.

TI The ***Mycobacterium*** tuberculosis complex-restricted gene cfp32 encodes an expressed protein that is detectable in tuberculosis patients and is positively correlated with pulmonary. . .

AB Human tuberculosis (TB) is caused by the bacillus ***Mycobacterium*** tuberculosis, a subspecies of the M. tuberculosis complex (MTC) of mycobacteria. Postgenomic dissection of the M. tuberculosis proteome is ongoing. . . CF and cytosolic compartments. Data to support the in vivo expression of CFP32 were provided by the serum recognition of ***recombinant*** CFP32 in 32% of TB patients by enzyme-linked immunosorbent assay (ELISA) as well as the direct detection of CFP32 by. . .

STP KeyWords Plus (R): CULTURE FILTRATE ANTIGENS; T-CELL RESPONSES; ANTIBODY-RESPONSES; SUPEROXIDE-DISMUTASE; ***GLUTAMINE*** - ***SYNTHETASE*** ; DISEASE PROGRESSION; PROTECTIVE IMMUNITY; GEL-ELECTROPHORESIS; CYTOKINE PRODUCTION; GENOMIC DELETIONS

L10 ANSWER 19 OF 32 SCISEARCH COPYRIGHT (c) 2009 The Thomson Corporation on STN

AN 2003:763661 SCISEARCH <<LOGINID::20090416>>

GA The Genuine Article (R) Number: 716NC

TI Purification and biochemical characterization of ***recombinant*** ***alanine*** ***dehydrogenase*** from Thermus caldophilus GK24

AU Shin H J (Reprint)

CS EnzBank Inc, KRIBB, BVC, Taejon 305333, South Korea (Reprint)

AU Bae J D; Cho Y J; Kim D I; Lee D S

CS KRIBB, Mol Glycobiol Res Unit, Taejon 305333, South Korea

CYA South Korea

SO JOURNAL OF MICROBIOLOGY AND BIOTECHNOLOGY, (AUG 2003) Vol. 13, No. 4, pp. 628-631.
ISSN: 1017-7825.

PB KOREAN SOC MICROBIOLOGY & BIOTECHNOLOGY, KOREA SCI TECHNOL CENTER #507, 635-4 YEOGSAM-DONG, KANGNAM-GU, SEOUL 135-703, SOUTH KOREA.

DT Article; Journal

LA English

REC Reference Count: 25

ED Entered STN: 19 Sep 2003
Last Updated on STN: 19 Sep 2003

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB The ***recombinant*** ***alanine*** ***dehydrogenase*** (ADH) from E. coli containing Thermus caldophilus ADH was purified to homogeneity from a cell-free extract. The enzyme was purified 38-fold

with a yield of 68% from the starting cell-free extract. The purified enzyme gave a single band in polyacrylamide gel electrophoresis, and its molecular weight was estimated to be 45 kDa. The pH optimum was 8.0 for reductive amination of pyruvate and 12.0 for oxidative deamination of L-alanine. The enzyme was stable up to 70degreesC. The activity of the enzyme was inhibited by 1 mM Zn2+ , 20% hexane, and 20% CHCl3. However, 10 mM Mg2+ and 40% propanol had no effect on the enzyme activity. The Michaelis constants (K-m) for the substrates were 50 muM for NADH, 0.2 mM for pyruvate, 39.4 mM for NH4+, 2.6 mM for Lalanine, and 1.8 mM for NAD(+).

TI Purification and biochemical characterization of ***recombinant*** ***alanine*** ***dehydrogenase*** from Thermus caldophilus GK24

AB The ***recombinant*** ***alanine*** ***dehydrogenase*** (ADH) from E. coli containing Thermus caldophilus ADH was purified to homogeneity from a cell-free extract. The enzyme was purified. . .

ST Author Keywords: ***alanine*** ***dehydrogenase*** ; characterization; enzyme purification; Thermus caldophilus GK24

STP KeyWords Plus (R): ***MYCOBACTERIUM*** -TUBERCULOSIS; BACILLUS-SUBTILIS; CLONING; GENE; EXPRESSION; METABOLISM; MECHANISM; STRAINS; ANTIGEN; ENZYME

L10 ANSWER 20 OF 32 BIOSIS COPYRIGHT (c) 2009 The Thomson Corporation on STN DUPLICATE 8

AN 2002:600488 BIOSIS <<LOGINID::20090416>>

DN PREV200200600488

TI Production of avirulent mutants of ***Mycobacterium*** bovis with vaccine properties by the use of illegitimate recombination and screening of stationary-phase cultures.

AU Collins, D. M. [Reprint author]; Wilson, T.; Campbell, S.; Buddle, B. M.; Wards, B. J.; Hotter, G.; De Lisle, G. W.

CS Wallaceville Animal Research Centre, AgResearch, PO Box 40063, Upper Hutt, New Zealand
desmond.collins@agresearch.co.nz

SO Microbiology (Reading), (October, 2002) Vol. 148, No. 10, pp. 3019-3027. print.
ISSN: 1350-0872.

DT Article

LA English

ED Entered STN: 20 Nov 2002
Last Updated on STN: 20 Nov 2002

AB A better tuberculosis vaccine is urgently required to control the continuing epidemic. Molecular techniques are now available to produce a better live vaccine than BCG by producing avirulent strains of the ***Mycobacterium*** tuberculosis complex with known gene deletions. In this study, 1000 illegitimate ***recombinants*** of ***Mycobacterium*** bovis were produced by illegitimate recombination with fragments of mycobacterial DNA containing a kanamycin resistance gene. Eight ***recombinant*** strains were selected on the basis of their inability to grow when stationary-phase cultures were inoculated into minimal medium. Five of these ***recombinants*** were found to be avirulent when inoculated into guinea pigs. Two of the avirulent ***recombinants*** produced vaccine efficacy comparable to BCG against an aerosol challenge in guinea pigs with M. bovis. One of these ***recombinants*** had an inactivated glnA2 gene encoding a putative ***glutamine*** ***synthetase*** . Transcriptional analysis showed that inactivation of glnA2 did not affect expression of the downstream glnE gene. The other ***recombinant*** had a block of 12 genes

deleted, including the sigma factor gene sigG. Two avirulent ***recombinants*** with an inactivated pckA gene, encoding phosphoenolpyruvate carboxykinase which catalyses the first step of gluconeogenesis, induced poor protection against tuberculosis. It is clear that live avirulent strains of the M. tuberculosis complex vary widely in their ability as vaccines to protect against tuberculosis. Improved models may be required to more clearly determine the difference in protective effect between BCG and potential new tuberculosis vaccines.

TI Production of avirulent mutants of ***Mycobacterium*** bovis with vaccine properties by the use of illegitimate recombination and screening of stationary-phase cultures.

AB. . . epidemic. Molecular techniques are now available to produce a better live vaccine than BCG by producing avirulent strains of the ***Mycobacterium*** tuberculosis complex with known gene deletions. In this study, 1000 illegitimate ***recombinants*** of ***Mycobacterium*** bovis were produced by illegitimate recombination with fragments of mycobacterial DNA containing a kanamycin resistance gene. Eight ***recombinant*** strains were selected on the basis of their inability to grow when stationary-phase cultures were inoculated into minimal medium. Five of these ***recombinants*** were found to be avirulent when inoculated into guinea pigs. Two of the avirulent ***recombinants*** produced vaccine efficacy comparable to BCG against an aerosol challenge in guinea pigs with M. bovis. One of these ***recombinants*** had an inactivated glnA2 gene encoding a putative ***glutamine*** ***synthetase*** . Transcriptional analysis showed that inactivation of glnA2 did not affect expression of the downstream glnE gene. The other ***recombinant*** had a block of 12 genes deleted, including the sigma factor gene sigG. Two avirulent ***recombinants*** with an inactivated pckA gene, encoding phosphoenolpyruvate carboxykinase which catalyses the first step of gluconeogenesis, induced poor protection against tuberculosis.. . .

IT . . .
Methods and Techniques; Molecular Genetics (Biochemistry and Molecular Biophysics); Pharmacology

IT Diseases
tuberculosis: bacterial disease
Tuberculosis (MeSH)

IT Chemicals & Biochemicals
Mycobacterium bovis vaccine: immunologic-drug, immunostimulant-drug, vaccine; ***glutamine*** ***synthetase***

ORGN . . .
Humans, Mammals, Primates, Vertebrates

ORGN Classifier
Mycobacteriaceae 08881

Super Taxa
Mycobacteria; Actinomycetes and Related Organisms; Eubacteria; Bacteria; Microorganisms

Organism Name
Mycobacterium bovis: avirulent
Mycobacterium tuberculosis: pathogen

Taxa Notes
Bacteria, Eubacteria, Microorganisms

RN 9023-70-5 (***glutamine*** ***synthetase***)

GEN ***Mycobacterium*** bovis glnA2 gene (Mycobacteriaceae);
Mycobacterium bovis sigG gene (Mycobacteriaceae)

L10 ANSWER 21 OF 32 BIOSIS COPYRIGHT (c) 2009 The Thomson Corporation on

STN DUPLICATE 9

AN 2001:504088 BIOSIS <<LOGINID::20090416>>

DN PREV200100504088

TI High extracellular levels of ***Mycobacterium*** tuberculosis ***glutamine*** ***synthetase*** and superoxide dismutase in actively growing cultures are due to high expression and extracellular stability rather than to a protein-specific export mechanism.

AU Tullius, Michael V.; Harth, Gunter; Horwitz, Marcus A. [Reprint author]

CS Division of Infectious Diseases, Department of Medicine, School of Medicine, UCLA, 10833 Le Conte Ave., CHS 37-121, Los Angeles, CA, 90095-1688, USA
mhorwitz@mednet.ucla.edu

SO Infection and Immunity, (October, 2001) Vol. 69, No. 10, pp. 6348-6363. print.
CODEN: INFIBR. ISSN: 0019-9567.

DT Article

LA English

OS Genbank-AF061031; Genbank-AY008693

ED Entered STN: 31 Oct 2001
Last Updated on STN: 25 Feb 2002

AB ***Glutamine*** ***synthetase*** (GS) and superoxide dismutase (SOD), large multimeric enzymes that are thought to play important roles in the pathogenicity of ***Mycobacterium*** tuberculosis, are among the bacterium's major culture filtrate proteins in actively growing cultures. Although these proteins lack a leader peptide, their presence in the extracellular medium during early stages of growth suggested that they might be actively secreted. To understand their mechanism of export, we cloned the homologous genes (glnA1 and sodA) from the rapid-growing, nonpathogenic ***Mycobacterium*** smegmatis, generated glnA1 and sodA mutants of M. smegmatis by allelic exchange, and quantitated expression and export of both mycobacterial and nonmycobacterial GSs and SODs in these mutants. We also quantitated expression and export of homologous and heterologous SODs from M. tuberculosis. When each of the genes was expressed from a multicopy plasmid, M. smegmatis exported comparable proportions of both the M. tuberculosis and M. smegmatis GSs (in the glnA1 strain) or SODs (in the sodA strain), in contrast to previous observations in wild-type strains. Surprisingly, ***recombinant*** M. smegmatis and M. tuberculosis strains even exported nonmycobacterial SODs. To determine the extent to which export of these large, leaderless proteins is expression dependent, we constructed a ***recombinant*** M. tuberculosis strain expressing green fluorescent protein (GFP) at high levels and a ***recombinant*** M. smegmatis strain coexpressing the M. smegmatis GS, M. smegmatis SOD, and M. tuberculosis BfrB (bacterioferritin) at high levels. The ***recombinant*** M. tuberculosis strain exported GFP even in early stages of growth and at proportions very similar to those of the endogenous M. tuberculosis GS and SOD. Similarly, the ***recombinant*** M. smegmatis strain exported bacterioferritin, a large (apprx500-kDa), leaderless, multimeric protein, in proportions comparable to GS and SOD. In contrast, high-level expression of the large, leaderless, multimeric protein malate dehydrogenase did not lead to extracellular accumulation because the protein was highly unstable extracellularly. These findings indicate that, contrary to expectations, export of M. tuberculosis GS and SOD in actively growing cultures is not due to a protein-specific export mechanism, but rather to bacterial leakage or autolysis, and that the extracellular abundance of these enzymes is simply due to their high level of expression and extracellular stability. The same determinants likely

explain the presence of other leaderless proteins in the extracellular medium of actively growing M. tuberculosis cultures.

TI High extracellular levels of ***Mycobacterium*** tuberculosis ***glutamine*** ***synthetase*** and superoxide dismutase in actively growing cultures are due to high expression and extracellular stability rather than to a protein-specific. . .

AB ***Glutamine*** ***synthetase*** (GS) and superoxide dismutase (SOD), large multimeric enzymes that are thought to play important roles in the pathogenicity of ***Mycobacterium*** tuberculosis, are among the bacterium's major culture filtrate proteins in actively growing cultures. Although these proteins lack a leader peptide,. . . actively secreted. To understand their mechanism of export, we cloned the homologous genes (glnA1 and sodA) from the rapid-growing, nonpathogenic ***Mycobacterium*** smegmatis, generated glnA1 and sodA mutants of M. smegmatis by allelic exchange, and quantitated expression and export of both mycobacterial. . . GSs (in the glnA1 strain) or SODs (in the sodA strain), in contrast to previous observations in wild-type strains. Surprisingly, ***recombinant*** M. smegmatis and M. tuberculosis strains even exported nonmycobacterial SODs. To determine the extent to which export of these large, leaderless proteins is expression dependent, we constructed a ***recombinant*** M. tuberculosis strain expressing green fluorescent protein (GFP) at high levels and a ***recombinant*** M. smegmatis strain coexpressing the M. smegmatis GS, M. smegmatis SOD, and M. tuberculosis BfrB (bacterioferritin) at high levels. The ***recombinant*** M. tuberculosis strain exported GFP even in early stages of growth and at proportions very similar to those of the endogenous M. tuberculosis GS and SOD. Similarly, the ***recombinant*** M. smegmatis strain exported bacterioferritin, a large (apprx500-kDa), leaderless, multimeric protein, in proportions comparable to GS and SOD. In contrast,. . .

IT Major Concepts
Cell Biology; Molecular Genetics (Biochemistry and Molecular Biophysics)

IT Chemicals & Biochemicals
glutamine ***synthetase*** [GS]: expression, extracellular stability, leaderless; malate dehydrogenase: multimeric protein; superoxide dismutase [SOD]: expression, extracellular stability, leaderless

ORGN . . .
Notes
Bacteria, Eubacteria, Microorganisms

ORGN Classifier
Mycobacteriaceae 08881

Super Taxa
Mycobacteria; Actinomycetes and Related Organisms; Eubacteria; Bacteria; Microorganisms

Organism Name
Mycobacterium smegmatis: gene expression system, strain-1-2c
Mycobacterium tuberculosis: strain-ATCC 35801

Taxa Notes
Bacteria, Eubacteria, Microorganisms

RN 9023-70-5 (***glutamine*** ***synthetase***)
9023-70-5 (GS)
9001-64-3 (malate dehydrogenase)
9054-89-1 (superoxide dismutase)
9054-89-1 (SOD)
222619-19-4 (Genbank-AF061031)

360028-71-3 (Genbank-AY008693)

GEN ***Mycobacterium*** tuberculosis glnA1 gene (Mycobacteriaceae): mutant; ***Mycobacterium*** tuberculosis sodA gene (Mycobacteriaceae): mutant

L10 ANSWER 22 OF 32 BIOSIS COPYRIGHT (c) 2009 The Thomson Corporation on STN DUPLICATE 10

AN 2000:103072 BIOSIS <<LOGINID::20090416>>

DN PREV200000103072

TI Treatment of ***Mycobacterium*** tuberculosis with antisense oligonucleotides to ***glutamine*** ***synthetase*** mRNA inhibits ***glutamine*** ***synthetase*** activity, formation of the poly-L-glutamate/glutamine cell wall structure, and bacterial replication.

AU Harth, Gunter; Zamecnik, Paul C.; Tang, Jin-Yan; Tabatadze, David; Horwitz, Marcus A. [Reprint author]

CS Division of Infectious Diseases, Department of Medicine, School of Medicine, University of California, 10833 Le Conte Avenue, 37-121 CHS, Los Angeles, CA, 90095, USA

SO Proceedings of the National Academy of Sciences of the United States of America, (Jan. 4, 2000) Vol. 97, No. 1, pp. 418-423. print. CODEN: PNASA6. ISSN: 0027-8424.

DT Article

LA English

ED Entered STN: 22 Mar 2000
Last Updated on STN: 3 Jan 2002

AB New antibiotics to combat the emerging pandemic of drug-resistant strains of ***Mycobacterium*** tuberculosis are urgently needed. We have investigated the effects on M. tuberculosis of phosphorothioate-modified antisense oligodeoxyribonucleotides (PS-ODNs) against the mRNA of ***glutamine*** ***synthetase*** , an enzyme whose export is associated with pathogenicity and with the formation of a poly-L-glutamate/glutamine cell wall structure. Treatment of virulent M. tuberculosis with 10 muM antisense PS-ODNs reduced ***glutamine*** ***synthetase*** activity and expression by 25-50% depending on whether one, two, or three different PS-ODNs were used and the PS-ODNs' specific target sites on the mRNA. Treatment with PS-ODNs of a ***recombinant*** strain of ***Mycobacterium*** smegmatis expressing M. tuberculosis ***glutamine*** ***synthetase*** selectively inhibited the ***recombinant*** enzyme but not the endogenous enzyme for which the mRNA transcript was mismatched by 2-4 nt. Treatment of M. tuberculosis with the antisense PS-ODNs also reduced the amount of poly-L-glutamate/glutamine in the cell wall by 24%. Finally, treatment with antisense PS-ODNs reduced M. tuberculosis growth by 0.7 logs (1 PS-ODN) to 1.25 logs (3 PS-ODNs) but had no effect on the growth of M. smegmatis, which does not export ***glutamine*** ***synthetase*** nor possess the poly-L-glutamate/glutamine (P-L-glx) cell wall structure. The experiments indicate that the antisense PS-ODNs enter the cytoplasm of M. tuberculosis and bind to their cognate targets. Although more potent ODN technology is needed, this study demonstrates the feasibility of using antisense ODNs in the antibiotic armamentarium against M. tuberculosis.

TI Treatment of ***Mycobacterium*** tuberculosis with antisense oligonucleotides to ***glutamine*** ***synthetase*** mRNA inhibits ***glutamine*** ***synthetase*** activity, formation of the poly-L-glutamate/glutamine cell wall structure, and bacterial replication.

AB New antibiotics to combat the emerging pandemic of drug-resistant strains of ***Mycobacterium*** tuberculosis are urgently needed. We have investigated the effects on M. tuberculosis of phosphorothioate-modified

antisense oligodeoxyribonucleotides (PS-ODNs) against the mRNA of ***glutamine*** ***synthetase*** , an enzyme whose export is associated with pathogenicity and with the formation of a poly-L-glutamate/glutamine cell wall structure. Treatment of virulent M. tuberculosis with 10 muM antisense PS-ODNs reduced ***glutamine*** ***synthetase*** activity and expression by 25-50% depending on whether one, two, or three different PS-ODNs were used and the PS-ODNs' specific target sites on the mRNA. Treatment with PS-ODNs of a ***recombinant*** strain of ***Mycobacterium*** smegmatis expressing M. tuberculosis ***glutamine*** ***synthetase*** selectively inhibited the ***recombinant*** enzyme but not the endogenous enzyme for which the mRNA transcript was mismatched by 2-4 nt. Treatment of M. tuberculosis. . . PS-ODN) to 1.25 logs (3 PS-ODNs) but had no effect on the growth of M. smegmatis, which does not export ***glutamine*** ***synthetase*** nor possess the poly-L-glutamate/glutamine (P-L-glx) cell wall structure. The experiments indicate that the antisense PS-ODNs enter the cytoplasm of M.. . .

IT Major Concepts
Biochemistry and Molecular Biophysics; Infection

IT Chemicals & Biochemicals
glutamine ***synthetase*** : activity inhibition, expression; ***glutamine*** ***synthetase*** mRNA; mRNA; phosphorothioate-modified antisense oligodeoxyribonucleotides; poly-L-glutamate/glutamine: cell wall structure formation

ORGN Classifier
Mycobacteriaceae 08881

Super Taxa
Mycobacteria; Actinomycetes and Related Organisms; Eubacteria; Bacteria; Microorganisms

Organism Name
Mycobacterium smegmatis: pathogen
Mycobacterium tuberculosis: pathogen, replication, virulent

Taxa Notes
Bacteria, Eubacteria, Microorganisms

RN 9023-70-5 (***glutamine*** ***synthetase***)

L10 ANSWER 23 OF 32 BIOSIS COPYRIGHT (c) 2009 The Thomson Corporation on STN DUPLICATE 11

AN 2000:106001 BIOSIS <<LOGINID::20090416>>

DN PREV200000106001

TI Evaluation of ***Mycobacterium*** tuberculosis genes involved in resistance to killing by human macrophages.

AU Miller, Barbara H.; Shinnick, Thomas M. [Reprint author]

CS Centers for Disease Control and Prevention, 1600 Clifton Rd., Atlanta, GA, 30329, USA

SO Infection and Immunity, (Jan., 2000) Vol. 68, No. 1, pp. 387-390. print. CODEN: INFIBR. ISSN: 0019-9567.

DT Article

LA English

ED Entered STN: 22 Mar 2000
Last Updated on STN: 3 Jan 2002

AB A coinfection assay was developed to examine ***Mycobacterium*** tuberculosis genes suspected to be involved in resistance to killing by human macrophages. THP-1 macrophages were infected with a mixture of equal numbers of ***recombinant*** ***Mycobacterium*** smegmatis LR222 bacteria expressing an M. tuberculosis gene and wild-type M. smegmatis LR222 bacteria expressing the xylE gene. At various times after

infection, the infected macrophages were lysed and the bacteria were plated. The resulting colonies were sprayed with catechol to determine the number of ***recombinant*** colonies and the number of xylE-expressing colonies. M. smegmatis bacteria expressing the M. tuberculosis ***glutamine*** ***synthetase*** A (glnA) gene or open reading frame Rv2962c or Rv2958c demonstrated significantly increased survival rates in THP-1 macrophages relative to those of xylE-expressing bacteria. M. smegmatis bacteria expressing M. tuberculosis genes for phospholipase C (plcA and plcB) or for high temperature requirement A (htrA) did not.

TI Evaluation of ***Mycobacterium*** tuberculosis genes involved in resistance to killing by human macrophages.

AB A coinfection assay was developed to examine ***Mycobacterium*** tuberculosis genes suspected to be involved in resistance to killing by human macrophages. THP-1 macrophages were infected with a mixture of equal numbers of ***recombinant*** ***Mycobacterium*** smegmatis LR222 bacteria expressing an M. tuberculosis gene and wild-type M. smegmatis LR222 bacteria expressing the xylE gene. At various. . . macrophages were lysed and the bacteria were plated. The resulting colonies were sprayed with catechol to determine the number of ***recombinant*** colonies and the number of xylE-expressing colonies. M. smegmatis bacteria expressing the M. tuberculosis ***glutamine*** ***synthetase*** A (glnA) gene or open reading frame Rv2962c or Rv2958c demonstrated significantly increased survival rates in THP-1 macrophages relative to. . .

IT . . .
and Homeostasis); Infection

IT Parts, Structures, & Systems of Organisms
macrophages: blood and lymphatics, immune system

IT Chemicals & Biochemicals
Mycobacterium ***glutamine*** ***synthetase*** A gene; ***Mycobacterium*** phospholipase C gene; ***Mycobacterium*** xylE gene

ORGN . . .
Humans, Mammals, Primates, Vertebrates

ORGN Classifier
Mycobacteriaceae 08881

Super Taxa
Mycobacteria; Actinomycetes and Related Organisms; Eubacteria; Bacteria; Microorganisms

Organism Name
Mycobacterium smegmatis: pathogen
Mycobacterium tuberculosis: pathogen

Taxa Notes
Bacteria, Eubacteria, Microorganisms

L10 ANSWER 24 OF 32 SCISEARCH COPYRIGHT (c) 2009 The Thomson Corporation on STN

AN 1999:144332 SCISEARCH <<LOGINID::20090416>>

GA The Genuine Article (R) Number: 166KQ

TI Export of ***recombinant*** ***Mycobacterium*** tuberculosis superoxide dismutase is dependent upon both information in the protein and mycobacterial export machinery - A model for studying export of leaderless proteins by pathogenic mycobacteria

AU Horwitz M A (Reprint)

CS Univ Calif Los Angeles, Sch Med, Dept Med, Div Infect Dis, 10833 Le Conte Ave, Los Angeles, CA 90095 USA (Reprint)

AU Harth G
CS Univ Calif Los Angeles, Sch Med, Dept Med, Div Infect Dis, Los Angeles, CA 90095 USA
CYA USA
SO JOURNAL OF BIOLOGICAL CHEMISTRY, (12 FEB 1999) Vol. 274, No. 7, pp. 4281-4292.
ISSN: 0021-9258.
PB AMER SOC BIOCHEMISTRY MOLECULAR BIOLOGY INC, 9650 ROCKVILLE PIKE, BETHESDA, MD 20814 USA.
DT Article; Journal
LA English
REC Reference Count: 28
ED Entered STN: 1999
Last Updated on STN: 1999
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS
AB We have investigated the expression and extracellular release of enzymatically active superoxide dismutase, one of the 10 major extracellular proteins of ***Mycobacterium*** tuberculosis, both in its native host and in the heterologous host ***Mycobacterium*** smegmatis, We found that the M, tuberculosis superoxide dismutase gene, encoding a leaderless polypeptide of M-r similar to 23,000 representing one of the four identical subunits of the enzyme, is expressed constitutively under normal growth conditions and at a B-fold increased level under conditions of hydrogen peroxide stress. The highly pathogenic ***mycobacterium*** M. tuberculosis expresses 93-fold more superoxide dismutase than the nonpathogenic ***mycobacterium*** M. smegmatis, and it exports a much higher proportion of expressed enzyme (76 versus 21%); taking both expression and export into consideration, IM, tuberculosis exports similar to 350-fold more enzyme than M, smegmatis, In M. smegmatis, ***recombinant*** M. tuberculosis superoxide dismutase is expressed at 8.4 times the level of the endogenous enzyme and the proportion exported (66%) approaches that in the homologous host; hence M. smegmatis exports up to 26-fold more of the ***recombinant*** than endogenous enzyme. Interestingly, subunits of the M. tuberculosis and M. smegmatis enzymes readily and stoichiometrically exchange with each other, forming five different complexes of four subunits, both when the enzymes are expressed in the ***recombinant*** host and when the purified enzymes are incubated together; however, each subunit retains its characteristic metal ion, iron for IM. tuberculosis and manganese for M.. smegmatis, Compared with the cell-associated enzyme, the supernatant enzyme of ***recombinant*** IM, smegmatis is enriched for M, tuberculosis enzyme subunits, consistent with preferential export of the M. tuberculosis enzyme. ***Recombinant*** M. tuberculosis superoxide dismutase transcomplements a superoxide dismutase-deficient Escherichia coli, resulting in a reduction of sensitivity of the strain to oxidative stress, but the enzyme is not exported from this nonmycobacterial host. Our findings indicate that the information for export of the M, tuberculosis superoxide dismutase is contained within the protein but that export additionally requires export machinery specific to mycobacteria.
TI Export of ***recombinant*** ***Mycobacterium*** tuberculosis superoxide dismutase is dependent upon both information in the protein and mycobacterial export machinery - A model for studying. . .
AB . . . have investigated the expression and extracellular release of enzymatically active superoxide dismutase, one of the 10 major extracellular proteins of ***Mycobacterium*** tuberculosis, both in its native host and in the heterologous host ***Mycobacterium*** smegmatis, We found that the M, tuberculosis superoxide dismutase gene,

encoding a leaderless polypeptide of M-r similar to 23,000 representing. . . constitutively under normal growth conditions and at a B-fold increased level under conditions of hydrogen peroxide stress. The highly pathogenic ***mycobacterium*** M. tuberculosis expresses 93-fold more superoxide dismutase than the nonpathogenic ***mycobacterium*** M. smegmatis, and it exports a much higher proportion of expressed enzyme (76 versus 21%); taking both expression and export into consideration, IM, tuberculosis exports similar to 350-fold more enzyme than M, smegmatis, In M. smegmatis, ***recombinant*** M. tuberculosis superoxide dismutase is expressed at 8.4 times the level of the endogenous enzyme and the proportion exported (66%) approaches that in the homologous host; hence M. smegmatis exports up to 26-fold more of the ***recombinant*** than endogenous enzyme. Interestingly, subunits of the M. tuberculosis and M. smegmatis enzymes readily and stoichiometrically exchange with each other, forming five different complexes of four subunits, both when the enzymes are expressed in the ***recombinant*** host and when the purified enzymes are incubated together; however, each subunit retains its characteristic metal ion, iron for IM. tuberculosis and manganese for M.. smegmatis, Compared with the cell-associated enzyme, the supernatant enzyme of ***recombinant*** IM, smegmatis is enriched for M, tuberculosis enzyme subunits, consistent with preferential export of the M. tuberculosis enzyme. ***Recombinant*** M. tuberculosis superoxide dismutase transcomplements a superoxide dismutase-deficient Escherichia coli, resulting in a reduction of sensitivity of the strain to. . .

STP KeyWords Plus (R): ESCHERICHIA-COLI; ***GLUTAMINE*** - ***SYNTHETASE*** ; EXPRESSION; GENE; IDENTIFICATION; PHAGOCYTOSIS; RECEPTORS; SEQUENCE; ANTIGEN; RELEASE

L10 ANSWER 25 OF 32 BIOSIS COPYRIGHT (c) 2009 The Thomson Corporation on STN DUPLICATE 12

AN 1999:241758 BIOSIS <<LOGINID::20090416>>

DN PREV199900241758

TI Preliminary crystallographic studies on ***glutamine*** ***synthetase*** from ***Mycobacterium*** tuberculosis.

AU Gill, Harindarpal S.; Pfluegl, Gaston M. U.; Eisenberg, David [Reprint author]

CS Departments of Chemistry and Biochemistry and Biological Chemistry, UCLA-DOE Laboratory of Structural Biology and Molecular Medicine, University of California Los Angeles, Los Angeles, CA, 90095-1570, USA

SO Acta Crystallographica Section D Biological Crystallography, (April, 1999) Vol. 55, No. 4, pp. 865-868. print.
ISSN: 0907-4449.

DT Article

LA English

ED Entered STN: 17 Jun 1999
Last Updated on STN: 17 Jun 1999

AB The etiologic agent of tuberculosis, ***Mycobacterium*** tuberculosis, has been shown to secrete the enzyme ***glutamine*** ***synthetase*** (TB-GS) which is apparently essential for infection. Four crystal forms of a ***recombinant*** TB-GS were grown. The one chosen for synchrotron X-ray data collection belongs to space group P212121 with unit-cell dimensions 208 X 258 X 274 ANG, yielding 2.4 ANG resolution data. A Matthews number of 2.89 ANG3 Da-1 is found, corresponding to 24 subunits of molecular mass 1300 kDa in the asymmetric unit. From earlier work, the structure of Salmonella typhimurium GS, which is 51% identical in sequence to TB-GS, is known to be dodecameric with 622 symmetry. Self-rotation calculations on the TB-GS X-ray data

reveal only one set of sixfold and twofold axes of symmetry. A Patterson map calculated from the native X-ray data confirms that there are two dodecamers in the asymmetric unit, having both their sixfold and twofold axes parallel to one another.

TI Preliminary crystallographic studies on ***glutamine*** ***synthetase*** from ***Mycobacterium*** tuberculosis.

AB The etiologic agent of tuberculosis, ***Mycobacterium*** tuberculosis, has been shown to secrete the enzyme ***glutamine*** ***synthetase*** (TB-GS) which is apparently essential for infection. Four crystal forms of a ***recombinant*** TB-GS were grown. The one chosen for synchrotron X-ray data collection belongs to space group P212121 with unit-cell dimensions 208. . .

IT Major Concepts
Enzymology (Biochemistry and Molecular Biophysics); Methods and Techniques

IT Chemicals & Biochemicals
glutamine ***synthetase*** : characterization, ***recombinant*** , structure

IT Methods & Equipment
hanging drop vapor-diffusion crystallization: chemical modification, sample preparation method, crystallization techniques; ***recombinant*** protein protocol: synthesis/modification techniques, synthetic method; X-ray crystallography: X-ray analysis, analytical method

ORGN . . .

Notes
Bacteria, Eubacteria, Microorganisms

ORGN Classifier
Mycobacteriaceae 08881

Super Taxa
Mycobacteria; Actinomycetes and Related Organisms; Eubacteria; Bacteria; Microorganisms

Organism Name
Mycobacterium tuberculosis

Taxa Notes
Bacteria, Eubacteria, Microorganisms

RN 9023-70-5 (***glutamine*** ***synthetase***)

L10 ANSWER 26 OF 32 BIOSIS COPYRIGHT (c) 2009 The Thomson Corporation on STN DUPLICATE 13

AN 2000:26849 BIOSIS <<LOGINID::20090416>>

DN PREV200000026849

TI Properties of the 40 kDa antigen of ***Mycobacterium*** tuberculosis, a functional L- ***alanine*** ***dehydrogenase*** .

AU Hutter, Bernd; Singh, Mahavir [Reprint author]

CS GBF (Gesellschaft fuer Biotechnologische Forschung m.b.H)-National Research Center for Biotechnology and Department of Biochemistry, Technical University of Braunschweig, 38124, Braunschweig, Germany

SO Biochemical Journal, (Nov. 1, 1999) Vol. 343, No. 3, pp. 669-672. print. ISSN: 0264-6021.

DT Article

LA English

ED Entered STN: 13 Jan 2000
Last Updated on STN: 31 Dec 2001

AB The 40 kDa antigen of ***Mycobacterium*** tuberculosis is the first antigen reported to be present in the pathogenic M. tuberculosis, but not in the vaccine strain ***Mycobacterium*** bovis BCG. It is a

functional L- ***alanine*** ***dehydrogenase*** (EC 1.4.1.1) and hence one of the few antigens possessing an enzymic activity. This makes the 40 kDa antigen attractive for potential diagnostic and therapeutic interventions. Recently, we developed a strategy to purify quantities of the ***recombinant*** protein in active form, and here we describe the biochemical properties of this enzyme. In the oxidative-deamination reaction, the enzyme showed Km values of 13.8 mM and 0.31 mM for L-alanine and NAD+, respectively, in a random-ordered mechanism. Km,app values in the reductive-amination reaction are 35.4 mM, 1.45 mM and 98.2 muM for ammonium, pyruvate and NADH, respectively. The enzyme is highly specific for all of its substrates in both directions. The pH profile indicates that oxidative deamination virtually may not occur at physiological pH. Hence L-alanine most likely is the product of the reaction catalysed in vivo. The enzyme is heat-stable, losing practically no activity at 60 degreeC for several hours.

TI Properties of the 40 kDa antigen of ***Mycobacterium*** tuberculosis, a functional L- ***alanine*** ***dehydrogenase*** .

AB The 40 kDa antigen of ***Mycobacterium*** tuberculosis is the first antigen reported to be present in the pathogenic M. tuberculosis, but not in the vaccine strain ***Mycobacterium*** bovis BCG. It is a functional L- ***alanine*** ***dehydrogenase*** (EC 1.4.1.1) and hence one of the few antigens possessing an enzymic activity. This makes the 40 kDa antigen attractive for potential diagnostic and therapeutic interventions. Recently, we developed a strategy to purify quantities of the ***recombinant*** protein in active form, and here we describe the biochemical properties of this enzyme. In the oxidative-deamination reaction, the enzyme. . .

IT Major Concepts
Enzymology (Biochemistry and Molecular Biophysics)

IT Chemicals & Biochemicals
Mycobacterium tuberculosis L- ***alanine*** ***dehydrogenase*** [EC 1.4.1.1]: 40 kDa antigen

L10 ANSWER 27 OF 32 CAPLUS COPYRIGHT 2009 ACS on STN
AN 1998:79429 CAPLUS <<LOGINID::20090416>>
DN 128:151095
OREF 128:29677a,29680a
TI Cloning of gene for NAD+-dependent formate dehydrogenase from ***Mycobacterium*** vaccae and use for the enzymic preparation of amino acids in presence of NADH-dependent amino acid dehydrogenase
IN Sauta, Kenji; Esaki, Nobuyoshi; Galkin, Andre
PA Unitika Ltd., Japan
SO Jpn. Kokai Tokkyo Koho, 13 pp.
CODEN: JKXXAF
DT Patent
LA Japanese
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI JP 10023896	A	19980127	JP 1996-217060	19960819
PRAI JP 1996-112303	A	19960507		

AB A ***recombinant*** plasmid encoding NAD+-dependent formate dehydrogenase (I) of ***Mycobacterium*** vaccae strain S10 and an NADH-dependent amino acid dehydrogenase is prepd. for transformation of Escherichia coli. The transgenic Escherichia coli is then used for the prodn. of amino acids via coupled reactions of the 2 enzymes in the presence of .alpha.-keto acids and ammonium formate. Plasmid pFDH/LeuDH

encoding I and leucine dehydrogenase of Thermoactinomyces intermedicus was prepd. and used for the transformation of E. coli. The transgenic E. coli was able to efficiently produce L-leucine from .alpha.-keto-isocaproic acid.

TI Cloning of gene for NAD+-dependent formate dehydrogenase from ***Mycobacterium*** vaccae and use for the enzymic preparation of amino acids in presence of NADH-dependent amino acid dehydrogenase

AB A ***recombinant*** plasmid encoding NAD+-dependent formate dehydrogenase (I) of ***Mycobacterium*** vaccae strain S10 and an NADH-dependent amino acid dehydrogenase is prepd. for transformation of Escherichia coli. The transgenic Escherichia coli. . .

ST ***Mycobacterium*** formate dehydrogenase gene sequence; Escherichia transgenic amino acid dehydrogenase; leucine prepn transgenic Escherichia

IT Escherichia coli
Fermentation
(cloning of gene for NAD+-dependent formate dehydrogenase from ***Mycobacterium*** vaccae and use for enzymic prepn. of amino acids in presence of NADH-dependent amino acid dehydrogenase)

IT Gene, microbial
RL: BOC (Biological occurrence); BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study); OCCU (Occurrence)
(cloning of gene for NAD+-dependent formate dehydrogenase of ***Mycobacterium*** vaccae)

IT DNA sequences
(for NAD+-dependent formate dehydrogenase of ***Mycobacterium*** vaccae)

IT Molecular cloning
(gene for NAD+-dependent formate dehydrogenase of ***Mycobacterium*** vaccae)

IT Protein sequences
(of NAD+-dependent formate dehydrogenase of ***Mycobacterium*** vaccae)

IT Carboxylic acids, reactions
RL: RCT (Reactant); RACT (Reactant or reagent)
(oxo; cloning of gene for NAD+-dependent formate dehydrogenase from ***Mycobacterium*** vaccae and use for enzymic prepn. of amino acids in presence of NADH-dependent amino acid dehydrogenase)

IT 202758-71-2
RL: BAC (Biological activity or effector, except adverse); BSU (Biological study, unclassified); BUU (Biological use, unclassified); PRP (Properties); BIOL (Biological study); USES (Uses)
(amino acid sequence; cloning of gene for NAD+-dependent formate dehydrogenase from ***Mycobacterium*** vaccae and use for enzymic prepn. of amino acids in presence of NADH-dependent amino acid dehydrogenase)

IT 9029-06-5, ***Alanine*** ***dehydrogenase*** 9082-71-7, Leucine dehydrogenase 53414-75-8, Amino acid dehydrogenase 69403-12-9, Phenylalanine dehydrogenase
RL: BAC (Biological activity or effector, except adverse); BSU (Biological study, unclassified); BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(cloning of gene for NAD+-dependent formate dehydrogenase from ***Mycobacterium*** vaccae and use for enzymic prepn. of amino acids in presence of NADH-dependent amino acid dehydrogenase)

IT 56-41-7P, L-Alanine, preparation 61-90-5P, L-Leucine, preparation 63-68-3P, L-Methionine, preparation 63-91-2P, L-Phenylalanine, preparation 72-18-4P, L-Valine, preparation 327-57-1P, L-Norleucine

1492-24-6P, L-.alpha.-Aminobutyric acid 6600-40-4P, L-Norvaline
RL: BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation)
(cloning of gene for NAD+-dependent formate dehydrogenase from ***Mycobacterium*** vaccae and use for enzymic prepn. of amino acids in presence of NADH-dependent amino acid dehydrogenase)

IT 540-69-2, Ammonium formate 583-92-6 600-18-0, .alpha.-Ketobutyric acid 759-05-7, .alpha.-keto-Isovaleric acid 1821-02-9, .alpha.-Ketovaleric acid 2492-75-3
RL: RCT (Reactant); RACT (Reactant or reagent)
(cloning of gene for NAD+-dependent formate dehydrogenase from ***Mycobacterium*** vaccae and use for enzymic prepn. of amino acids in presence of NADH-dependent amino acid dehydrogenase)

IT 202758-70-1
RL: BOC (Biological occurrence); BSU (Biological study, unclassified); BUU (Biological use, unclassified); PRP (Properties); BIOL (Biological study); OCCU (Occurrence); USES (Uses)
(nucleotide sequence; cloning of gene for NAD+-dependent formate dehydrogenase from ***Mycobacterium*** vaccae and use for enzymic prepn. of amino acids in presence of NADH-dependent amino acid dehydrogenase)

L10 ANSWER 28 OF 32 MEDLINE on STN
AN 1998311074 MEDLINE <<LOGINID::20090416>>
DN PubMed ID: 9648740
TI Cloning of an EF-P homologue from Bacteroides fragilis that increases B. fragilis ***glutamine*** ***synthetase*** activity in Escherichia coli.
AU Abratt V R; Mbewe M; Woods D R
CS Department of Microbiology, University of Cape Town, Rondebosch, South Africa.. val@molbiol.uct.ac.za
SO Molecular & general genetics : MGG, (1998 May) Vol. 258, No. 4, pp. 363-72.
Journal code: 0125036. ISSN: 0026-8925.
CY GERMANY: Germany, Federal Republic of
DT Journal; Article; (JOURNAL ARTICLE)
(RESEARCH SUPPORT, NON-U.S. GOV'T)
LA English
FS Priority Journals
OS GENBANK-U75509
EM 199807
ED Entered STN: 31 Jul 1998
Last Updated on STN: 3 Mar 2000
Entered Medline: 20 Jul 1998
AB Investigations of possible regulators of Bacteroides fragilis ***glutamine*** ***synthetase*** (GS) activity were done in Escherichia coli using a compatible dual-plasmid system. The B. fragilis glnA gene, together with upstream and downstream flanking regions, was cloned onto the low copy number plasmid pACYC184 and expressed in the E. coli glnA ntrB ntrC deletion strain, YMC11. GS activity was monitored following co-transformation with a B. fragilis genomic library carried on the compatible plasmid pEcoR251. A gene was cloned that caused a twofold increase in B. fragilis GS activity but did not affect the activity of the E. coli GS enzyme or the B. fragilis sucrase (ScrL). Deletion of the B. fragilis glnA downstream region decreased basal levels of GS activity, but did not affect the ability of the cloned gene to increase the B. fragilis GS activity. Reporter gene analysis, using the B. fragilis glnA promoter

region fused to the promoterless Clostridium acetobutylicum endoglucanase gene, showed no increase in reporter gene activity. This demonstrated that the increase in GS activity was not regulated at the transcriptional level, and that the cloned gene product was not affecting the copy number of the plasmid in trans. Sequence data indicated that the cloned gene had good amino acid identity to a range of elongation factor P (EF-P) proteins, the highest being to that of a Synechocystis sp (48%), and the least to ***Mycobacterium*** genitalium (27%). Amino acid identity to the E. coli EF-P was intermediate (37%). A possible role for EF-P in enhancing translation of the B. fragilis glnA mRNA is proposed.

TI Cloning of an EF-P homologue from Bacteroides fragilis that increases B. fragilis ***glutamine*** ***synthetase*** activity in Escherichia coli.

AB Investigations of possible regulators of Bacteroides fragilis ***glutamine*** ***synthetase*** (GS) activity were done in Escherichia coli using a compatible dual-plasmid system. The B. fragilis glnA gene, together with upstream. . . of elongation factor P (EF-P) proteins, the highest being to that of a Synechocystis sp (48%), and the least to ***Mycobacterium*** genitalium (27%). Amino acid identity to the E. coli EF-P was intermediate (37%). A possible role for EF-P in enhancing. . .

CT . . . ME, metabolism
Molecular Sequence Data
*Peptide Elongation Factors: GE, genetics
Peptide Elongation Factors: ME, metabolism
Promoter Regions, Genetic
Protein Biosynthesis
*** Recombinant Proteins: GE, genetics***
*** Recombinant Proteins: ME, metabolism***
Transfection

CN 0 (DNA, Bacterial); 0 (Peptide Elongation Factors); 0 (***Recombinant*** Proteins); 0 (factor EF-P); EC 6.3.1.2 (Glutamate-Ammonia Ligase)

L10 ANSWER 29 OF 32 BIOSIS COPYRIGHT (c) 2009 The Thomson Corporation on STN DUPLICATE 14

AN 1998:317427 BIOSIS <<LOGINID::20090416>>

DN PREV199800317427

TI Host vector system for high-level expression and purification of ***recombinant*** , enzymatically active ***alanine*** ***dehydrogenase*** of ***Mycobacterium*** tuberculosis.

AU Hutter, Bernd; Singh, Mahavir [Reprint author]

CS GBF-German Natl. Res. Cent. Biotechnol., Mascheroder Weg 1, D-38123 Braunschweig, Germany

SO Gene (Amsterdam), (May 28, 1998) Vol. 212, No. 1, pp. 21-29. print. CODEN: GENED6. ISSN: 0378-1119.

DT Article

LA English

OS Genbank-U92472

ED Entered STN: 22 Jul 1998
Last Updated on STN: 22 Jul 1998

AB The 40-kDa antigen of M. tuberculosis, which is an ***alanine*** ***dehydrogenase*** , is a species-specific antigen that is potentially useful for strain identification. Large quantities of the purified protein are required for immunological, as well as for detailed biochemical and structural, characterization. The AlaDH gene was cloned by PCR from H37Rv (virulent) and H37Ra (partially attenuated) strains of M. tuberculosis, and their DNA sequence was determined. A host-vector

system suitable for the production of sufficient quantities of the ***recombinant*** AlaDH antigen was developed. The AlaDH gene was expressed under the control of strong, transcriptional (bacteriophage pLpR) and translational (atpE) signals. High-level expression of soluble AlaDH was obtained using the ***recombinant*** E. coli K-12 strain CAG629 (pMSK12), which is deficient in Lon protease and the heat-shock response. A simple two-step procedure for the rapid purification of the ***recombinant*** protein was developed. The, protein was purified to near homogeneity, and the purified AlaDH showed a specific enzyme activity comparable to the native protein isolated from M. tuberculosis. In addition, the product showed an expected amino acid sequence and reacted strongly to the 40-kDa (AlaDH)specific mAb HBT-10. Furthermore, the epitope of the mAb HBT-10 was mapped to a 12-amino-acid region. Contrary to the published results, we show that the AlaDH and the PNT (pyridine nucleotide transhydrogenase) of M. tuberculosis do not share common epitopes reacting to the species-specific mAb HBT-10. The availability of highly purified AlaDH should now enable a detailed biochemical and structural characterization of this important enzyme of M. tuberculosis.

TI Host vector system for high-level expression and purification of ***recombinant*** , enzymatically active ***alanine*** ***dehydrogenase*** of ***Mycobacterium*** tuberculosis.

AB The 40-kDa antigen of M. tuberculosis, which is an ***alanine*** ***dehydrogenase*** , is a species-specific antigen that is potentially useful for strain identification. Large quantities of the purified protein are required for. . . M. tuberculosis, and their DNA sequence was determined. A host-vector system suitable for the production of sufficient quantities of the ***recombinant*** AlaDH antigen was developed. The AlaDH gene was expressed under the control of strong, transcriptional (bacteriophage pLpR) and translational (atpE) signals. High-level expression of soluble AlaDH was obtained using the ***recombinant*** E. coli K-12 strain CAG629 (pMSK12), which is deficient in Lon protease and the heat-shock response. A simple two-step procedure for the rapid purification of the ***recombinant*** protein was developed. The, protein was purified to near homogeneity, and the purified AlaDH showed a specific enzyme activity comparable. . .

IT Major Concepts
Enzymology (Biochemistry and Molecular Biophysics); Molecular Genetics (Biochemistry and Molecular Biophysics)

IT Chemicals & Biochemicals
alanine ***dehydrogenase*** : antigen, characterization, purification, expression; pyridine nucleotide transhydrogenase; AlaDH gene: cloning; DNA: extraction, sequencing

ORGN . . .
Notes
Bacteria, Eubacteria, Microorganisms
ORGN Classifier
Mycobacteriaceae 08881
Super Taxa
Mycobacteria; Actinomycetes and Related Organisms; Eubacteria; Bacteria; Microorganisms
Organism Name
Mycobacterium -tuberculosis: strain-H37Ra, strain-H37Rv
Taxa Notes
Bacteria, Eubacteria, Microorganisms

RN 9029-06-5 (***alanine*** ***dehydrogenase***)
9014-18-0 (pyridine nucleotide transhydrogenase)

L10 ANSWER 30 OF 32 BIOSIS COPYRIGHT (c) 2009 The Thomson Corporation on STN DUPLICATE 15

AN 1997:460535 BIOSIS <<LOGINID::20090416>>

DN PREV199799759738

TI Expression and efficient export of enzymatically active ***Mycobacterium*** tuberculosis ***glutamine*** ***synthetase*** in ***Mycobacterium*** smegmatis and evidence that the information for export is contained within the protein.

AU Harth, Gunter; Horwitz, Marcus A. [Reprint author]

CS Div. Infect. Dis., Dep. Med., 37-121 CHS, Sch. Med., UCLA, 10883 Le Conte Ave., Los Angeles, CA 90095, USA

SO Journal of Biological Chemistry, (1997) Vol. 272, No. 36, pp. 22728-22735. CODEN: JBCHA3. ISSN: 0021-9258.

DT Article

LA English

ED Entered STN: 27 Oct 1997
Last Updated on STN: 27 Oct 1997

AB We have investigated the expression and extracellular release of active, ***recombinant*** ***Mycobacterium*** tuberculosis ***glutamine*** ***synthetase*** (EC 6.3.1.2), an enzyme that is a potentially important determinant of M. tuberculosis infection and whose extracellular release is correlated with pathogenicity. The M. tuberculosis ***glutamine*** ***synthetase*** gene encodes a polypeptide of 478 amino acids; 12 such subunits comprise the active enzyme. Northern blot, nuclease S1, and primer extension analyses revealed ***glutamine*** ***synthetase*** specific transcripts of apprx 1,550 and 1,650 nucleotides produced under low and high nitrogen conditions, respectively. Expression of ***recombinant*** M. tuberculosis ***glutamine*** ***synthetase*** in Escherichia coli YMC21E, a ***glutamine*** ***synthetase*** deletion mutant, led to transcomplementation of the mutant but not to release of active enzyme. Expression in ***Mycobacterium*** smegmatis 1-2c, from the gene's own promoter, resulted in the release of gt 95% of all ***recombinant*** enzyme. No hybrid molecules containing M. tuberculosis and M. smegmatis ***glutamine*** ***synthetase*** subunits were detected. Native and ***recombinant*** exported and intracellular ***glutamine*** ***synthetase*** molecules were indistinguishable from one another by mass, N-terminal amino acid sequence, antibody reactivity, and enzymatic activity. Since M. tuberculosis ***glutamine*** ***synthetase*** is similar to other, strictly intracellular, bacterial glutamine synthetases and the DNA sequence upstream of the structural gene does not encode a leader peptide, the information to target the protein for export must be contained in its amino acid sequence and/or conformation.

TI Expression and efficient export of enzymatically active ***Mycobacterium*** tuberculosis ***glutamine*** ***synthetase*** in ***Mycobacterium*** smegmatis and evidence that the information for export is contained within the protein.

AB We have investigated the expression and extracellular release of active, ***recombinant*** ***Mycobacterium*** tuberculosis ***glutamine*** ***synthetase*** (EC 6.3.1.2), an enzyme that is a potentially important determinant of M. tuberculosis infection and whose extracellular release is correlated with pathogenicity. The M. tuberculosis ***glutamine***

synthetase gene encodes a polypeptide of 478 amino acids; 12 such subunits comprise the active enzyme. Northern blot, nuclease S1, and primer extension analyses revealed ***glutamine*** ***synthetase*** specific transcripts of apprx 1,550 and 1,650 nucleotides produced under low and high nitrogen conditions, respectively. Expression of ***recombinant*** M. tuberculosis ***glutamine*** ***synthetase*** in Escherichia coli YMC21E, a ***glutamine*** ***synthetase*** deletion mutant, led to transcomplementation of the mutant but not to release of active enzyme. Expression in ***Mycobacterium*** smegmatis 1-2c, from the gene's own promoter, resulted in the release of gt 95% of all ***recombinant*** enzyme. No hybrid molecules containing M. tuberculosis and M. smegmatis ***glutamine*** ***synthetase*** subunits were detected. Native and ***recombinant*** exported and intracellular ***glutamine*** ***synthetase*** molecules were indistinguishable from one another by mass, N-terminal amino acid sequence, antibody reactivity, and enzymatic activity. Since M. tuberculosis ***glutamine*** ***synthetase*** is similar to other, strictly intracellular, bacterial glutamine synthetases and the DNA sequence upstream of the structural gene does not. . .

IT Major Concepts
Biochemistry and Molecular Biophysics; Enzymology (Biochemistry and Molecular Biophysics); Genetics; Physiology

IT Chemicals & Biochemicals
GLUTAMINE ***SYNTHETASE*** ; EC 6.3.1.2

IT Miscellaneous Descriptors
EC 6.3.1.2; ENZYMOLOGY; EXPORT; EXPRESSION; GENOMIC ORGANIZATION; ***GLUTAMINE*** ***SYNTHETASE*** ; ***GLUTAMINE*** ***SYNTHETASE*** GENE; MOLECULAR GENETICS; PRODUCTION; ***RECOMBINANT*** ENZYME; U87280

ORGN . . .
Notes
Bacteria, Eubacteria, Microorganisms

ORGN Classifier
Mycobacteriaceae 08881

Super Taxa
Mycobacteria; Actinomycetes and Related Organisms; Eubacteria; Bacteria; Microorganisms

Organism Name
Mycobacterium smegmatis
Mycobacterium tuberculosis

Taxa Notes
Bacteria, Eubacteria, Microorganisms

RN 9023-70-5 (***GLUTAMINE*** ***SYNTHETASE***)
9023-70-5 (EC 6.3.1.2)

L10 ANSWER 31 OF 32 BIOSIS COPYRIGHT (c) 2009 The Thomson Corporation on STN DUPLICATE 16

AN 1997:86799 BIOSIS <<LOGINID::20090416>>
DN PREV199799378512
TI Use of rpsL for dominance selection and gene replacement in Streptomyces roseosporus.
AU Hosted, Thomas J.; Baltz, Richard H. [Reprint author]
CS Lilly Res. Lab., A Div. Eli Lilly Company, Lilly Corporate Cent., Indianapolis, IN 46258-0424, USA
SO Journal of Bacteriology, (1997) Vol. 179, No. 1, pp. 180-186.
CODEN: JOBAAY. ISSN: 0021-9193.

DT Article
LA English
OS Genbank-U60191
ED Entered STN: 26 Feb 1997
Last Updated on STN: 2 Apr 1997
AB We developed a gene replacement system using the rpsL gene of Streptomyces roseosporus and demonstrated its utility by constructing a deletion in the S. roseosporus glnA gene. A 1.3-kb BamHI fragment that hybridized to the ***Mycobacterium*** smegmatis rpsL gene was subcloned from an S. roseosporus cosmid library and sequenced. Plasmid pRHB514 containing the rpsL gene conferred streptomycin sensitivity (Sm-s) to the Sm-r S. roseosporus TH149. The temperature-sensitive plasmid pRHB543 containing rpsL and the S. roseosporus glnA gene disrupted with a hygromycin resistance (Hm-r) gene was introduced into S. roseosporus TH149, and ***recombinants*** containing single and double crossovers were obtained after a temperature increase. Southern hybridization analysis revealed that single crossovers occurred in the glnA or rpsL genes and that double crossovers resulted in replacement of the chromosomal glnA gene with the disrupted glnA. ***Glutamine*** ***synthetase*** activity was undetectable in the ***recombinant*** containing the disrupted glnA gene.
AB. . . its utility by constructing a deletion in the S. roseosporus glnA gene. A 1.3-kb BamHI fragment that hybridized to the ***Mycobacterium*** smegmatis rpsL gene was subcloned from an S. roseosporus cosmid library and sequenced. Plasmid pRHB514 containing the rpsL gene conferred. . . and the S. roseosporus glnA gene disrupted with a hygromycin resistance (Hm-r) gene was introduced into S. roseosporus TH149, and ***recombinants*** containing single and double crossovers were obtained after a temperature increase. Southern hybridization analysis revealed that single crossovers occurred in. . . glnA or rpsL genes and that double crossovers resulted in replacement of the chromosomal glnA gene with the disrupted glnA. ***Glutamine*** ***synthetase*** activity was undetectable in the ***recombinant*** containing the disrupted glnA gene.
IT . . .
Enzymology (Biochemistry and Molecular Biophysics); Genetics; Metabolism; Molecular Genetics (Biochemistry and Molecular Biophysics); Physiology
IT Chemicals & Biochemicals
STREPTOMYCIN; HYGROMYCIN; ***GLUTAMINE*** ***SYNTHETASE***
IT . . .
Descriptors
ANALYTICAL METHOD; CHROMOSOME; COSMID LIBRARY; CROSSOVERS; DOMINANCE SELECTION; E.-COLI STRAIN-S17-1; E.-COLI STRAIN-XL1-BLUE MFR; GENE DELETIONS; GENE REPLACEMENT; GLNA GENE; ***GLUTAMINE*** ***SYNTHETASE*** ; HYGROMYCIN RESISTANCE GENE; MOLECULAR GENETICS; PLASMID PRHB514; PLASMID PRHB543; RPSL GENE; SOUTHERN HYBRIDIZATION; STREPTOMYCIN SENSITIVITY; TEMPERATURE SENSITIVITY
ORGN . . .
Notes
Bacteria, Eubacteria, Microorganisms
ORGN Classifier
Mycobacteriaceae 08881
Super Taxa
Mycobacteria; Actinomycetes and Related Organisms; Eubacteria; Bacteria; Microorganisms

Organism Name
Mycobacterium smegmatis
Taxa Notes
Bacteria, Eubacteria, Microorganisms
ORGN Classifier
Streptomycetes and Related Genera 08840
Super Taxa
Actinomycetes and Related Organisms; Eubacteria;. . .
RN 57-92-1 (STREPTOMYCIN)
6379-56-2 (HYGROMYCIN)
9023-70-5 (***GLUTAMINE*** ***SYNTHETASE***)
183640-69-9 (Genbank-U60191)

L10 ANSWER 32 OF 32 BIOSIS COPYRIGHT (c) 2009 The Thomson Corporation on STN DUPLICATE 17
AN 1997:248587 BIOSIS <<LOGINID::20090416>>
DN PREV199799547790
TI A study of combined filtration and adsorption on nylon-based dye-affinity membranes: Separation of ***recombinant*** L- ***alanine*** ***dehydrogenase*** from crude fermentation broth.
AU Weissenborn, Michael; Hutter, Bernd; Singh, Mahavir; Beeskow, Thomas C.; Anspach, F. Birger [Reprint author]
CS Biochem. Eng. Div., GBF, Gesellschaft Biotechnologische Forschung m.b.H., Mascheroder Weg 1, D-38124 Braunschweig, Germany
SO Biotechnology and Applied Biochemistry, (1997) Vol. 25, No. 2, pp. 159-168.
CODEN: BABIEC. ISSN: 0885-4513.
DT Article
LA English
ED Entered STN: 13 Jun 1997
Last Updated on STN: 13 Jun 1997
AB Dextran, hydroxyethylcellulose (HEC), and poly(vinyl alcohol) (PVA) were covalently linked to bisoxirane-activated nylon membranes. Cibacron Blue F3G-A was immobilized on to these membranes to yield a dye-affinity membrane. The hydrodynamic permeability of affinity membranes was reduced to apprxeq 50% of that of the original Nylon membrane due to extension of polymer coils into flow-through pores. Adsorption of pre-purified human serum albumin (HSA) and malate dehydrogenase (MDH) displayed highest maximum binding capacities on HEC-coated dye-ligand-affinity membranes, ranging from 163 mu-g/cm-2 for HSA to 316 mu-g/cm-2 for MDH. The protein recovery of HSA was 100% on dextran-coated membranes compared with 70% on PVA-coated membranes, whereas almost 100% recovery was found for MDH, independent of the polymer. Application of crude supernatant from ***recombinant*** Escherichia coli yielded purification factors of 7.4, 8.9 and 11.2 for ***recombinant*** ***alanine*** ***dehydrogenase*** from ***Mycobacterium*** tuberculosis for HEC-, dextran- and PVA-coated membranes respectively. Dynamic capacities decreased remarkably to apprxeq 3 mu-g/cm-2 to co-adsorption of host proteins. The presence of cell debris caused only a slight decrease of purification factors, but a dramatic decrease of the permeability of affinity membranes due to development of a particle layer in front of the membranes. Although enzyme recoveries were up to 90% using cell-free supernatant, more than 50% of the product was lost due to polarization, concentration and rejection at particle layers when using crude homogenates. In order to further improve this integrated downstream process, sophisticated membrane techniques are required by which the formation of a filter cake is circumvented. Further refinement of

polymer-coated membranes would not help one to avoid this problem.

TI A study of combined filtration and adsorption on nylon-based dye-affinity membranes: Separation of ***recombinant*** L- ***alanine*** ***dehydrogenase*** from crude fermentation broth.

AB. . . on PVA-coated membranes, whereas almost 100% recovery was found for MDH, independent of the polymer. Application of crude supernatant from ***recombinant*** Escherichia coli yielded purification factors of 7.4, 8.9 and 11.2 for ***recombinant*** ***alanine*** ***dehydrogenase*** from ***Mycobacterium*** tuberculosis for HEC-, dextran- and PVA-coated membranes respectively. Dynamic capacities decreased remarkably to apprxeq 3 mu-g/cm-2 to co-adsorption of host. .

.

IT . . .

Biophysics; Bioprocess Engineering; Enzymology (Biochemistry and Molecular Biophysics); Membranes (Cell Biology); Metabolism; Methods and Techniques; Physiology

IT Chemicals & Biochemicals

L- ***ALANINE*** ***DEHYDROGENASE*** ; DEXTRAN; HYDROXYETHYLCELLULOSE; POLY(VINYLALCOHOL); MALATE DEHYDROGENASE

IT Miscellaneous Descriptors

BIOBUSINESS; BIOPROCESS ENGINEERING; BIOTECHNOLOGY; DEXTRAN; ENZYMOLOGY; HUMAN SERUM ALBUMIN; HYDRODYNAMIC PERMEABILITY; HYDROXYETHYLCELLULOSE; INTEGRATED DOWNSTREAM PROCESS; L- ***ALANINE*** ***DEHYDROGENASE*** ; MALATE DEHYDROGENASE; MEMBRANE ADSORPTION; MEMBRANE FILTRATION; METHODOLOGY; NYLON-BASED DYE-AFFINITY MEMBRANES; POLY(VINYLALCOHOL); PURIFICATION METHOD; ***RECOMBINANT*** FORM; SEPARATION

ORGN . . .

microorganism

Taxa Notes

Microorganisms

ORGN Classifier

Mycobacteriaceae 08881

Super Taxa

Mycobacteria; Actinomycetes and Related Organisms; Eubacteria; Bacteria; Microorganisms

Organism Name

Mycobacterium tuberculosis

Taxa Notes

Bacteria, Eubacteria, Microorganisms

RN 9029-06-5 (L- ***ALANINE*** ***DEHYDROGENASE***)

9004-54-0 (DEXTRAN)

9004-62-0 (HYDROXYETHYLCELLULOSE)

9002-89-5 (POLY(VINYLALCOHOL))

9001-64-3 (MALATE DEHYDROGENASE)